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RENAL LESIONS ASSOCIATED WITH MULTIPLE MYELOMA*

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Proteinuria occurs in the great majority of persons afflicted with multiple myeloma, especially in the advanced stages of the disease, and according to the recent literature renal insufficiency is frequently demonstrable toward the end of the illness. In the postmortem reports that have been published the authors nearly all call attention to some renal lesion, but it would appear that there is no lesion peculiar to multiple myeloma. The purpose of this investigation is to determine the structural alterations in the kidneys responsible for proteinuria and renal insufficiency.

The protein in the urine may consist entirely of the Bence-Jones body, entirely of serum protein, or of a mixture of the two proteins. The more careful investigations indicate that when the Bence-Jones body is present it constitutes the greater part, or all of the urinary protein. The Bence-Jones protein is more frequently found in advanced than in early stages of the disease. In some instances the daily excretion of the protein remains fairly constant over long periods; in other instances it varies in quantity and may be absent for long or short intervals. It is recognized that errors may be made in testing for the Bence-Jones body. It is usually concluded that any protein that does not redissolve on boiling is serum protein, but Hewitt states that Bence-Jones proteins differ in their properties and are not all soluble in boiling solutions. Magnus-Levy states that when a large quantity of protein is found in the urine in a case of myeloma it is very probably Bence-Jones protein chiefly.

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It is difficult to determine accurately the percentage of cases of myeloma in which Bence-Jones proteinuria does not occur at any time. Obviously a negative report is not convincing unless the patient is followed to the end of the illness. Magnus-Levy, in 1932, from a careful survey of the literature concluded that about 73 per cent of the cases show Bence-Jones proteinuria. Geschickter and Copeland gave a somewhat lower estimate — 65 per cent.

The reports summarized in Table I include only cases in which the author gave information concerning renal function, blood pressure, or the histological structure of the kidneys. Reports that give no data on any of these points were not tabulated. The arrangement is in chronological order.

Myeloma without Proteinuria: Four cases have been reported in which no protein was found in the urine (Cases 8, 21, 22, 48). In Geschickter and Copeland's case (Case 48) the patient was living at the time of the report, and it is therefore possible that proteinuria developed subsequently. In the other 3 reports it is not stated how often the urine was examined, or how long before death the examination was made. Microscopic examination of the kidneys was not reported in any of the 3 cases, and no functional studies were made. We do not know, therefore, whether or not there are any functional or structural alterations in the kidneys in cases of myeloma where the urine is normal, but it is probable that no serious lesion is present, since only a slight injury is required to produce albuminuria.

Myeloma with Serum Albuminuria but without Bence-Jones Proteinuria: Another group which may be studied separately is that in which there was serum albumin but no Bence-Jones protein in the urine. Eight cases of this type have been reported (Cases 3, 12, 36, 43, 49, 50, 59, 74) and 2 of our cases fall into this category. In 4 of these only a trace of serum albumin was present, and in another albumin was sometimes absent and sometimes present in small amounts. In 2 instances there was heavy albuminuria. The functional studies on this group are inadequate. In Case 59 the concentration test (specific gravity 1004 to 1012) indicates impaired kidney function. In Perla and Hutner's case (Case 74) the postmortem report shows clearly a high degree of renal insufficiency. This is the only instance in the literature of proved renal insufficiency in a myeloma without Bence-Jones proteinuria. The authors

interpret the lesion as a nephrotic contracted kidney. We shall discuss this case more fully in a subsequent paragraph.

There are 5 other postmortem reports in this group. One showed pyelonephritis, another many casts with atrophy of the subcapsular zone, and 2 showed only slight changes of no significance. The diagnosis of chronic interstitial nephritis (Case 50) cannot be accurately interpreted. Probably it was an arteriosclerotic kidney.

The rather meager histological studies available indicate that the renal lesions are about the same in the group with only serum albumin in the urine as in those with Bence-Jones proteinuria. Two of our cases belong in this group.

CASE 80: A–30–75. A male, 70 years of age, was under observation the last 2 weeks of his life. The urine on 2 examinations showed casts and a small amount of albumin. Two tests for Bence-Jones protein gave negative results. There was a little residual urine due to hypertrophy of the prostate. The blood pressure was 152/82.

At postmortem the myeloma was found to involve the ribs and vertebrae. The kidneys weighed 180 gm. and 200 gm. each. There were a number of cysts in the cortices, the largest having a diameter of 3 cm., but the greater part of the cortical tissue was intact.

On microscopic examination there are no casts and no atrophic tubules. The glomeruli are normal except for a slight increase of endothelial nuclei.

CASE 81: A–32–691. A male, 59 years of age, was admitted to the hospital May 25, 1931. His symptoms began in May, 1930, and a diagnosis of multiple myeloma was established in October, 1930. Numerous examinations of the urine showed albumin from a faint trace to a small amount. Sometimes no albumin was demonstrable. Repeated careful examinations for Bence-Jones protein gave negative results. The hemoglobin fell from 85 per cent on admission to 69 per cent shortly before death. In June and July, 1930, the icteric index was high, 128 to 160 units, and there was bile in the urine. The blood calcium varied from 10.48 to 15.80 gm. per 100 cc. June 4, 1931, total serum protein 5.85 gm. — albumin 3.61 gm., globulin 2.24 gm. On April 9, 1932, the non-protein nitrogen was 41.6 mg. and the blood pressure was 132/82. There was no edema at any time. Death, April 20, 1932.

At postmortem the weight of the heart was 250 gm. The kidneys weighed 150 gm. and 160 gm. each.

On microscopic examination the large and medium sized arteries show marked intimal thickening. The arterioles are unaffected. There is a definite atrophy of the tubules and glomeruli in the outer

TABLE I

Summary of Reports from the Literature Dealing with Renal Changes Associated with Multiple Myeloma

Case No.	Author	Sex	Age	Proteinuria (no Bence-Jones test)	Serum albumin in urine	Bence-Jones proteinuria	Blood pressure	Phenolsulphonephthalein	Blood urea	Non-protein nitrogen	Urea nitrogen	Total serum protein	Serum albumin	Serum globulin	Kidneys
			yrs.					%	*mg.*	*mg.*	*mg.*	*gm.*	*gm.*	*gm.*	
1	Ellinger	M	45		?	+									A few casts
2	Jochmann and Schumm	F	37		?	+									Amyloid
3	Scheele and Herxheimer	M	50		+ +	−									Many casts, atrophy of subcapsular zone
4	Collins	M	56	+ −	?	?									Occasional hyaline glomeruli
5	Tschistowitsch and Kolessnikoff	F	36		?	+									Calcification
6	Hopkins and Savory	F	65		?	+									Slight interstitial fibrosis
7	Austin	M	72		+	+									Atrophy of subcapsular zone
8	Stumm	M	58		−	−									No microscopic
9	Boggs and Guthrie	M	64		−	+	180/?								No postmortem
10	Folin and Denis	M	39		−	+ + +									No postmortem
11	Groat and Brewer	M	37		?	+	135/?	20		81.9	40.8				No postmortem
12	Mieremet	F	57		tr	−									A few casts, small foci of lymphocytes
13	Vance	M	54		?	+									Slight chronic parenchymatous nephritis
14	Froboese	F	52	+											Pyelonephritis
15	Jacobson	M	63		−	+ +	116/60	0							Each kidney 7 gm., arteriosclerosis
16	Taylor, Miller, and Sweet				+	+									No postmortem
17	Rowe				?	+						6.8	4.8	2.0	No postmortem
18	Glaus	M	67		?	+	145/?								Calcification, amyloid

TABLE I (*continued*)

Case No.	Author	Sex	Age	Proteinuria (no Bence-Jones test)	Serum albumin in urine	Bence-Jones proteinuria	Blood pressure	Phenolsulphonephthalein	Blood urea	Non-protein nitrogen	Urea nitrogen	Total serum protein	Serum albumin	Serum globulin	Kidneys
			yrs.					%	*mg.*	*mg.*	*mg.*	*gm.*	*gm.*	*gm.*	
19	Pepper and Pearce	M	52		?	+ + +									Arteriosclerosis, casts, degeneration of tubules
20	Beck and McCleary	M	55		+	+									Many casts, chronic diffuse nephritis
21	Wallgren	M	58		−	−									No microscopic
22	"	F	65		−	−	150/?								No microscopic
23	"	M	51		+	+									No postmortem
24	"	M	67		?	+									Slight parenchymatous nephritis
25	"	F	67	+			160/?								No microscopic
26	"	F	50	+			160/?								No microscopic
27	"	F	63	+ −			105/?								Arteriosclerosis
28	Bloch	F	59		+	+ −									No postmortem
29	Thannhauser and Krauss				+	+	120/68			37.9 202.1		6.12 4.49			Nephrotic contracted kidneys
30	Walters	M	69		−	+	Nm.		Nm.	Nm.					(Living)
31	"	F	42		?	+	124/80 140/80	10 7	74.0	63.0					No postmortem
32	Löhlein	M	50		−	+									Casts, protein crystals, lymphocytic infiltration
33	Oftedal	M	41		?	+	128/96								No microscopic
34	Hansen	M	38		?	+	122/90								No postmortem
35	McConnell	M	49		?	+ + +	190/100								Fibrosis of cortex, atrophy of tubules, casts
36	Wood and Lucké	M	51		Tr.	−									Many hyaline glomeruli
37	Ellermann	M	48	+											Casts, fibrosis, lymphocytic infiltration
38	Stone	M	56		?	+	115/80			37.0					No microscopic
39	Aschner	F	46		+	+ + +	103/?								Embolic abscess

TABLE I (*continued*)

Case No	Author	Sex	Age	Proteinuria (no Bence-Jones test)	Serum albumin in urine	Bence-Jones proteinuria	Blood pressure	Phenolsulphonephthalein	Blood urea	Non-protein nitrogen	Urea nitrogen	Total serum protein	Serum albumin	Serum globulin	Kidneys
			yrs.					%	*mg.*	*mg.*	*mg.*	*gm.*	*gm.*	*gm.*	
40	Guggenheimer	M	54	+ ++ +			125/80		28.0						Leukemic infiltration, casts, arteriosclerosis
41	"	M	62		+	+	140/?		21.5						Arteriosclerosis, lymphocytic infiltration
42	Belden	F	58		+	+					5.0				(Living)
43	Meyerding	M	57		tr	−	150/80								No postmortem
44	Paul and Funk	M	42	+ +				65 24	48.0						Lymphocytes, casts, dilated tubules
45	Kreuzer	F	56	+			130/65								No microscopic
46	Charlton	F	52		+	+									Interstitial nephritis
47	Kleine	M	53		?	+	145/70								Many casts, severe parenchymatous and interstitial nephritis
48	Geschickter and Copeland	M	44		−	−		Nm.							(Living)
49	"	M	51		+ −	−	110/85								Pyelonephritis
50	"	F	71		+ + +	−	140/90								Chronic interstitial nephritis
51	"	M	66		?	+	104/62	50							Multiple abscesses
52	"	F	37		?	+	130/80								No microscopic
53	"	F	50		?	+									Chronic nephritis
54	"	M	62		?	+	158/108	30							(Living)
55	"	M	59		?	+		9 18		70.0 80.0					(Living)
56	"	M	55	+			135/80	40							Chronic diffuse nephritis
57	Perlzweig and others	M	40		?	+	175/100			34.0		12.32 13.84	1.42 4.06	10.11 9.09	(Living)
58	Marcovici	M	55		?	+ −	110/80								No postmortem
59	Schittenhelm	F	46		tr	−	180/85								(Living)

TABLE I (*continued*)

Case No.	Author	Sex	Age	Proteinuria (no Bence-Jones test)	Serum albumin in urine	Bence-Jones proteinuria	Blood pressure	Phenolsulphonephthalein	Blood urea	Non-protein nitrogen	Urea nitrogen	Total serum protein	Serum albumin	Serum globulin	Kidneys
			yrs.					%	*mg.*	*mg.*	*mg.*	*gm.*	*gm.*	*gm.*	
60	Bannick and Greene	M	47		+	+	110/60 145/100	5	218						(Living)
61	"	F	59		?	+	210/110	10 15	104 132	74.0		9.35	2.5	6.5	No postmortem
62	"	M	68		?	+	120/70		100 115						(Living)
63	"	M	49		?	+	145/85	15	84.0						No postmortem
64	"	M	43		?	+	125/45	62 66				6.5 7.3			(Living)
65	"	F	52		?	+	115/70	30	66.0						(Living)
66	"	M	60		?	+	175/115 210/140		62.0						(Living)
67	"	F	69		?	+	185/105	10	46.0						No postmortem
68	"	M	64		?	+	100/65	25		74.0		6.0			(Living)
69	"	M	56		?	+	140/80		50.0						(Living)
70	"	F	70		?	+	130/70	35							(Living)
71	"	M	55		?	+	140/85	45	33.0	40.0		10.08 10.75	4.0 4.45	6.08 6.09	(Living)
72	Hewitt	M	46		−	+		17 34	54.0 77.0			6.31	4.35	1.96	(Living)
73	"	F	66		−	+		55	30.0						(Living)
74	Perla and Hutner	M	71		+ + +	−	135/75		52.2						Casts, marked tubular atrophy
75	"	M	43		?	+ + +	130/80				64.0				Many casts
76	Magnus-Levy				−	+				35.0		6.8			(Living)
77	Mainzer (Ehrich)	M	46		−	+	140/95 165/95		14.8	46.9 89.0		7.38	4.69	2.21	Extensive tubular atrophy due to casts
78	Hallay and Odor	F	38		+	+ −	140/80			39.0					Arteriosclerosis, subacute glomerulonephritis
79	Buschke	M		+ + +			140/70 100/60		62.7 98.0						Many casts, lymphocytic foci

zone of the cortex near the capsule. This alteration is to be attributed to poor blood supply resulting from narrowing of the large arteries. It is frequently found in the senile kidney.

In all the cases in this group, except Perla and Hutner's, the renal lesions are readily explainable as the effects of arteriosclerosis or of complicating infections.

Bence-Jones Proteinuria but No Serum Albumin: In 9 cases the authors state clearly that the Bence-Jones body was the only protein present in the urine (Cases 9, 10, 15, 30, 32, 72, 73, 76, 77). In 3 of these (Cases 15, 72, 77) renal insufficiency was clearly established by functional tests; in 3 others (Cases 30, 73, 76) renal function was good at the time the test was made. Three postmortem reports are available. In Mainzer's case (Case 77) the anatomical basis of renal insufficiency was extensive obstruction of the tubules by casts — the obstruction resulting in extensive tubular atrophy. In Jacobson's case (Case 15) renal insufficiency was apparently due to arteriosclerosis. The kidneys weighed 75 gm. each, the arteries were thick-walled and there were many hyaline glomeruli. No importance was attached to casts. There were no functional studies in Löhlein's case (Case 32), but the tubules were obstructed by crystallized Bence-Jones protein. No doubt a number of other cases belong in this group, since many authors made no attempt to determine whether or not the urinary protein consisted entirely of Bence-Jones body.

In 15 of the tabulated reports, including 3 of our own, both Bence-Jones body and serum albumin were demonstrated in the urine. Usually the amount of serum albumin was very small in comparison with the large amount of Bence-Jones protein, *e. g.* Wright found 0.03 gm. of serum albumin and 0.33 gm. of Bence-Jones protein in 100 cc. of urine. Sometimes serum albumin is absent in most examinations, but present in small amount occasionally, as in 2 of our cases (Cases 82 and 83).

In 36 of the tabulated reports the Bence-Jones protein was demonstrated, but no test was made for serum albumin. Sixteen reports are tabulated, including 6 of our own, which contain some information of value, although the albuminous urine was not tested for the Bence-Jones protein.

Before proceeding to the general discussion the reports of our own cases will be given.

TABLE II

Summary of 11 Cases of Multiple Myeloma Reported in this Paper

Case No.	Autopsy No.	Sex	Age	Proteinuria (no Bence-Jones test)	Serum albumin in urine	Bence-Jones proteinuria	Blood pressure	Phenolsulphonephthalein	Non-protein nitrogen	Urea nitrogen	Total serum protein	Serum albumin	Serum globulin	Kidneys
			yrs.					%	*mg.*	*mg.*	*gm.*	*gm.*	*gm.*	
80	30–75	M	70		+	–	152/82							Slight increase of glomerular endothelium
81	32–691	M	59		– +	–	132/82		41.6		5.85	3.61	2.24	Arteriosclerosis
82	26–1096	M	54		– +	+	108/64			45.73				Casts with some tubular atrophy
83	32–930	M	51		– +	+	140/70	63 48	19.2 38.0		6.53	3.39	3.14	Casts with some tubular atrophy
84	31–408	F	38	+ +			100/60	5		50.0 58.8				Many casts with extensive tubular atrophy (Fig. 4)
85	32–872	M	51		+	+ +	130/80							Many casts, protein in glomerular capillaries
86	32–1024	M	52	+ + +					100 200					Protein in glomerular capillaries (Fig. 2)
87	29–1165	M	70	+										Arteriosclerosis, a few casts, protein in glomerular capillaries
88	30–110	M	47			+	115/82							Numerous casts, thickened basement membrane
89	31–311	M	74	+			130/60							Slight increase of glomerular endothelium
90	30–1207	M	74							95.0				Thickened basement membrane

CASE 82: A–26–1096. Male, 54 years of age, admitted to the hospital July 26, 1926. In 1920 a part of the sternum with portions of several ribs was removed because of a tumor of the sternum. This was not examined but was probably myeloma. After this operation he remained well for about 6 years. On admission he was poorly nourished, and complained of pain in the chest and shoulders. Roentgen-ray examination revealed multiple myeloma involving a large number of bones. The blood pressure was 108/64. July 28, blood urea nitrogen was 45.73 mg. Bence-Jones protein was found in the urine on each of 22 examinations. Usually it was the only protein present, but at times there was some serum albumin. There was no edema. Death, Dec. 13, 1926.

At postmortem the myeloma was demonstrated in the skull, ribs, clavicles, innominate bones and femurs. The heart weighed 300 gm. The right kidney weighed 205 gm. and the left 65 gm. The small kidney was covered with deep pits.

Microscopic sections through the pitted areas show wedge-shaped areas of atrophy. The peripheral portions of the tubules near the capsule are collapsed and atrophic, while the deeper portions near the medulla are distended with large casts. Some of the casts are surrounded by giant macrophages. The large atrophic areas that have largely replaced the cortex of the left kidney are clearly due to obstruction of the tubules by casts. The glomeruli in these atrophic areas show a simple atrophy but no hyalinization. In addition to the large atrophic areas there are occasional clusters of hyaline glomeruli with atrophic tubules near the capsular surface. These are apparently due to senile arteriosclerosis. The slight renal insufficiency in this case is therefore attributed to obstruction of the tubules by casts.

CASE 83: A–32–930. Male, 51 years of age, admitted to the hospital Nov. 9, 1931. Illness began in July, 1930, with pain in the right hip. In October, 1930, the pain had become so severe that he could not walk. A spontaneous fracture of the right hip occurred on Jan. 25, 1931. The fracture did not heal and the patient became permanently bedridden. Roentgen-ray examination on admission revealed widespread involvement of the bones with myeloma. Eleven consecutive examinations of the urine during his 7 months stay in the hospital showed the Bence-Jones protein on each occasion. Serum albumin was usually absent, but occasionally present in small amount. On Nov. 9, 1931, the blood pressure was 140/70; phenolsulphonephthalein 63 per cent (2 hours); nonprotein nitrogen 19.2 mg.; Rehberg test 103. On May 2, 1932, the non-protein nitrogen was 38 mg.; Rehberg test 31.7; blood calcium 18.2 gm.; total serum proteins 6.53 gm. — albumin 3.39 gm., globulin 3.14 gm.; phenolsulphonephthalein 48 per cent. Death, June 1, 1932.

The Rehberg test is low but the other tests indicate very slight functional disturbance.

At postmortem myeloma was found in the ribs, sternum, innominate bones, femurs, skull and mandible, and there were multiple nodules in the liver with a maximum diameter of 4.5 cm. There was no edema. The heart weighed 380 gm. The kidneys weighed 210 gm. and 250 gm. each. The external surfaces were smooth and the cortices on section were of a light reddish color.

Microscopic examination of the kidneys reveals numerous casts, chiefly in the convoluted tubules. Numerous small areas of tubular atrophy have resulted from obstruction of the tubules by the casts, but the great majority of the tubules are not atrophic. The majority of the glomerular capillaries show a definite irregular thickening of the basement membrane (Fig. 1), such as occurs in lipoid nephrosis and eclampsia (Bell). This alteration in the basement membrane is convincing evidence of injury of the capillary and is an anatomical basis for proteinuria.

CASE 84: A–31–408. Female, 38 years of age, admitted to the hospital March 3, 1931. In July, 1928, she first noticed dyspnea, edema of the legs and weakness. On Aug. 3, 1928, she delivered a full term infant. Edema persisted for several months after delivery and she never regained her former health. A diagnosis of multiple myeloma was established in March, 1929. In June, 1930, there was a spontaneous fracture of the left humerus. Albumin was found in the urine at this time. On admission there was a marked loss of weight, nausea, vomiting and malaise. The blood pressure was 100/60. The urine showed a heavy cloud of albumin. No test was made for Bence-Jones protein. The blood urea nitrogen was 50 mg. On March 6 the blood urea nitrogen was 58.8 mg., and there were clinical signs of intestinal obstruction. The phenolsulphonephthalein output was 5 per cent in two hours. Death, March 8, 1931.

At postmortem the intestinal obstruction was found to be due to extensive amyloidosis of the small intestine. The myeloma was widely distributed in the bones. There was no edema. The kidneys were somewhat reduced in size, and on section the cortices were cloudy.

Microscopic examination reveals enormous numbers of casts in the tubules. The portions of the tubules containing casts are greatly dilated but the distal portions near the capsule are collapsed and atrophic (Fig. 4). There are no changes in the glomeruli except an irregular thickening of the basement membrane.

CASE 85: A–32–872. Male, 51 years of age. Onset of illness in October, 1931, with spontaneous fracture of a rib. Admitted to the hospital April 3, 1932, complaining of extreme weakness, pain in the chest and poor appetite. Blood pressure 130/80. The urine contained a large amount of protein that was shown to

be mainly Bence-Jones. Roentgen-ray examination demonstrated myeloma in both humeri, the left scapula, the ribs and the vertebrae. The patient lost weight rapidly and died May 18, 1932.

At postmortem the myeloma was found widely distributed in the bones. There was no edema. The heart weighed 300 gm. The right kidney weighed 225 gm. and the left 300 gm. The external surfaces were smooth and the cortices were pale.

On microscopic examination numerous casts are found in both the convoluted and the collecting tubules. The casts are often partly surrounded by macrophages. In spite of the numerous casts there is very little tubular atrophy. One or two glomeruli in a section show a cast filling the capsular space, *i. e.* between the parietal and visceral epithelial layers. In a few glomeruli the capillaries are partly or completely filled with a precipitated protein such as is shown in Figures 2 and 3. This will be discussed subsequently. There are no convincing changes in the capillary basement membrane. No functional studies were made in this case, but the only alteration tending toward renal insufficiency was tubular obstruction by casts, and these were not present in sufficient numbers to cause a serious functional derangement.

CASE 86: A–32–1024.* The patient, a male 52 years of age, had an acute illness in January, 1931, which was diagnosed influenza by his family physician. He never recovered completely from this illness. His physician prescribed for him occasionally until October, 1931, when he advised extraction of his teeth because of anemia. Eleven teeth were extracted. Bleeding began immediately and continued in spite of all treatment. He had never bled excessively before. He was admitted to the hospital Nov. 9, 1931. The hemoglobin was 35 per cent; erythyrocytes 2,000,000; leukocytes 6600 — polymorphonuclears 76 per cent, lymphocytes 20 per cent, no immature leukocytes; platelets 298,000. The bleeding and clotting times were both markedly prolonged. He continued to bleed in spite of numerous transfusions. The blood calcium was normal. On November 11 the non-protein nitrogen was 100 mg.; creatinin, 2.9 mg. On November 17 the non-protein nitrogen was 200 mg.; uric acid 10 mg.; creatinin 5 mg. The urine showed continuously a heavy protein content, as well as numerous hyaline and granular casts. Erythrocytes were rarely seen. There was no test for Bence-Jones protein. Death, Nov. 17, 1931.

The bone marrow of the sternum, ribs, vertebrae, and femurs was hyperplastic and of a red color. Microscopically the growth proved to be a myeloma, but there was relatively little destruction of bone. The heart weighed 450 gm. There was a terminal edema of the lungs

* I am indebted to Dr. A. G. Foord for permission to publish this case.

with bronchopneumonia. The kidneys together weighed 380 gm. The external surfaces were smooth, the cortices on section soft and opaque.

Microscopically there are only a few casts in the collecting tubules. The tubules appear practically normal, but the glomeruli show a very unusual appearance (Fig. 2). The glomerular capillaries are partially or completely filled with a precipitated protein substance. All the capillaries in all the glomeruli are distended, but they contain very few erythrocytes, which is good evidence that they were filled in life with plasma having a high protein content. The fixing reagent (Zenker's fluid) precipitated the protein from solution as it precipitates protein from albuminous urine in the tubules. The protein of the plasma would naturally be more concentrated in the glomerular capillaries where filtration of water is taking place. The partial obstruction of the glomerular capillaries by a highly viscous plasma is offered as an explanation of the renal insufficiency from which the patient died. The substance in the capillaries is not as dense as amyloid. In this kidney an amyloid test was not applied, but in a similar instance (Case 87) the amyloid stain gave a negative result.

CASE 87: A–29–1165. Male, 70 years of age. Complained of pain in the right lumbar and right lower thoracic region. No careful clinical or laboratory studies were made. Albuminuria was continuously present. No test for Bence-Jones body.

At postmortem there was no edema. The heart weighed 375 gm., the kidneys together, 250 gm. The external surfaces of the kidneys were smooth.

On microscopic examination there is found a well defined, narrow, subcapsular zone of hyaline glomeruli and atrophic tubules that is obviously the result of arteriosclerosis. Elsewhere the tubules are normal. There are only a few casts. A few glomeruli show the capillaries distended with precipitated protein (Fig. 3) as described in the previous case. The substance in the capillaries reacted negatively to tests for amyloid.

CASE 88: A–30–110. Male, 47 years of age, admitted to the hospital Dec. 23, 1929, complaining of weakness, loss of weight, pain in the chest and shoulders, and headache. He had a chronic cough and was dyspneic on exertion. The pain in his chest and shoulders had been present for many months. The blood pressure was 115/82. A tumor mass was recognized in the sternum. The urine

contained a trace of protein and hyaline and granular casts. Death in coma, Jan. 13, 1930. A specimen of urine obtained postmortem showed Bence-Jones protein.

The postmortem revealed multiple myeloma. The heart weighed 355 gm. and was greatly dilated. The kidneys weighed 160 gm. and 165 gm. each. The external surfaces were smooth and the cortices pale.

Microscopic examination reveals numerous small casts in the convoluted tubules, many of which are partially calcified. Some giant macrophages are seen about the casts. The tubules are apparently not completely obstructed by the casts. There are numerous hyaline glomeruli, but the cause of their obliteration cannot be determined with certainty. There is a rather marked intimal thickening of the small arteries which may have caused obliteration of glomeruli. All the other glomeruli are of normal size, but they show a marked irregular thickening of the capillary basement membrane such as is shown in Figure 1.

CASE 89: A–31–311. Male, 74 years of age. His illness began with a severe backache following unusual exertion on Nov. 7, 1930. The pain continued to be intense and sometime in December, 1930, he developed numbness and difficulty in using his legs. On December 11 the urine contained a small amount of protein and the blood pressure was 130/60. No test for Bence-Jones protein was made. Later the lower extremities became paralyzed completely and a large bedsore developed over the sacrum. A diagnosis of "tumor compressing the spinal cord" was made. Death, Feb. 21, 1931.

The postmortem examination revealed a myeloma of the lamina of the eighth thoracic vertebra which had grown into the spinal canal and compressed the spinal cord. No other bones were involved. There was lobar pneumonia of the right lower lobe. The heart weighed 210 gm. The kidneys showed no gross evidences of disease.

On microscopic examination the only alteration is a slight increase of glomerular endothelium which was probably caused by the pneumonia.

CASE 90: A–30–1207. Male, 74 years of age, admitted to the hospital in coma. Death 2 hours later. No history could be obtained. The patient was emaciated and there was incontinence of urine and feces. Examination of the heart and lungs gave no positive findings. The blood urea nitrogen was 95 mg., and the blood sugar 0.082 per cent. No further studies were made.

At postmortem there was no edema. Myeloma of the ribs and sternum was demonstrated. The heart weighed 320 gm., the kidneys 115 gm. each.

Microscopically there are no casts, no atrophic tubules and no calcified structures. The capillary basement membrane in most of the glomeruli shows irregular thickenings such as are shown in Figure 1.

The clinical phenomena that suggest renal disease are albuminuria, hematuria, edema, hypertension and impaired renal function. To what extent are these symptoms and signs found in association with multiple myeloma?

Albuminuria: In rare instances there is no protein of any kind in the urine. Serum albumin may be found alone or with Bence-Jones protein, but it is absent frequently and seldom present in large amount. The urinary protein is chiefly Bence-Jones body in most instances when both proteins are present. The Bence-Jones protein may be excreted by apparently normal kidneys, and its presence does not prove a renal lesion. The presence of serum albumin, however, is satisfactory evidence of injury of the glomerular capillaries, but a very slight damage is sufficient to cause albuminuria and one should not make the diagnosis of nephritis in the clinical sense on the basis of albuminuria alone. In view of the small amount of serum albumin usually found and its frequent absence we should not expect to find a clinical nephritis with myeloma, but merely a mild renal injury.

Hematuria: None of the authors mentions hematuria in a case of myeloma, so that in this respect there is a distinction from glomerulonephritis.

Edema: Edema was present in only 6 out of the 90 cases surveyed. It is found chiefly in the lower extremities and is not prominent. It is probably attributable to malnutrition rather than to renal disease.

Hypertension: The blood pressure has been recorded in 54 of the tabulated reports (46 from the literature and 8 of our own). The highest systolic blood pressures were as follows: 100 to 119 mm. Hg., 12 cases; 120 to 139 mm. Hg., 16 cases; 140 to 149 mm. Hg., 12 cases; 150 to 159 mm. Hg., 4 cases; 160 to 169 mm. Hg., 3 cases; 170 to 179 mm. Hg., 1 case; 180 to 189 mm. Hg., 3 cases; 190 to 199 mm. Hg., 1 case; 200 to 210 mm. Hg., 2 cases. The systolic blood pressure was 150 mm. Hg. or above in fourteen instances — 26 per cent. This is about the incidence of primary hypertension in elderly persons, and therefore elevated blood pressure cannot be attributed to the effects of myeloma unless primary hypertension has been ex-

cluded. There is only 1 postmortem report on a subject with very high blood pressure (Case 35), and the kidneys were not described in detail in this instance. We do not know therefore what the renal lesions were in the cases with high blood pressure. There was a moderate terminal elevation in Mainzer's case (Case 77) in which the characteristic renal lesion of myeloma was pronounced.

Impaired Renal Function: Functional studies were made on 43 of the 90 cases under consideration. The tests are not all satisfactory but they indicate that renal function was normal in 12, slightly decreased in 7, and definitely decreased in 24 cases. It is probable that some of those with normal function would have shown impairment in a later stage of the disease, since renal insufficiency is usually a terminal phenomenon.

It may be concluded that renal insufficiency is common in the advanced stages of multiple myeloma, but in other clinical features there is little resemblance to glomerulonephritis. The absence of edema and the normal or increased serum proteins indicate that the renal lesion is not similar to that of lipoid nephrosis. Allard and Weber mentioned a lipemia in their case, but Perlzweig, Delrue and Geschickter found the blood cholesterol 130 mg. There are no other observations on the blood lipoids.

The Structural Changes in the Kidneys: It is commonly stated that a nephritis is found at postmortem in 80 per cent or more of cases of multiple myeloma, but a large variety of minor alterations have been called "parenchymatous" or "interstitial nephritis." The only conclusion justified by a study of the literature is that there is usually some lesion in the kidneys that may or may not be caused by the myeloma.

Discussion

1. We shall first consider the 24 cases in which moderate or severe renal insufficiency was demonstrated by functional tests. In this group 10 postmortem reports are available. In Jacobson's case the lesion was probably arteriosclerosis, since the author described small contracted kidneys with sclerotic arteries and hyaline glomeruli. This was presumably an accidental association of myeloma and arteriosclerosis; there is no evidence that the two diseases are related. Thannhauser and Krauss described the kidneys in a case of

myeloma in which death was due to uremia. The kidneys were small and there was widespread tubular atrophy with normal glomeruli. The authors interpreted the lesion as a nephrotic contracted kidney, and it has been widely accepted as such. However, Ehrich called attention to Bohnencamp's diagnosis of this case. Bohnencamp examined sections of these kidneys, which he obtained from Aschoff, and interpreted the tubular atrophy as secondary to obstruction of the tubules by casts. The implication is that Aschoff concurred in this opinion. The photomicrographs published by Thannhauser and Krauss show some normal tubules among the atrophic ones, which supports Bohnencamp's interpretation. It is highly probable, therefore, that the tubular atrophy in this widely discussed case was not primary but the result of obstruction by casts.

In Paul and Funk's patient the phenolsulphonephthalein excretion fell to 24 per cent, and the blood urea was 48 mg. There was a heavy cloud of albumin, but no test for the Bence-Jones protein was made. The kidneys were large and pale. There was a diffuse round cell infiltration of the cortex and medulla, and there were dilated tubules with many hyaline and cellular casts. The glomeruli were large and some showed adhesions to the tufts. Extensive deposits of calcium were found in the interstitial tissues of the cortex. The authors made a diagnosis of "subacute nephritis," but the description corresponds to tubular obstruction by casts, with an exudative interstitial nephritis (pyelonephritis).

Perla and Hutner in 1930 reported 2 cases with renal insufficiency:

(*a*) Male, 43 years of age; blood pressure 130/80; no edema; urea nitrogen 64 mg.; creatinin 5.5 mg.; calcium 14.6 mg.; erythrocytes 1,150,000; hemoglobin 15 per cent. The urine contained large amounts of albumin and of Bence-Jones protein, as well as granular casts. The concentration test gave a specific gravity from 1010 to 1011. At postmortem the kidneys weighed together 220 gm. The cortices on section were of a pale yellowish color. The glomeruli were for the most part intact. The cortical tubules were dilated and filled with casts. In the medulla there was an extensive degeneration of the tubules with replacement by connective tissue. Some tubules were calcified. The authors interpreted the lesion as "chiefly a severe nephrosis," yet the only alteration tending to cause renal insufficiency was obstruction of the tubules by casts.

(*b*) Male, 71 years of age; blood pressure 135/75; hemoglobin 20 per cent; erythrocytes 1,220,000; blood urea 52.2 mg.; creatinin 5.1 mg.; specific gravity 1012; large quantities of albumin in the urine but no Bence-Jones protein; no casts; no edema. The heart weighed 230 gm. Each kidney weighed 100 gm., the surfaces were smooth and the cortices narrow. Microscopically there was an enormous increase of interstitial tissue in the cortices with marked destruction, atrophy and replacement of tubules. The glomeruli were largely normal. The tubules were strikingly atrophic and some were dilated and filled with hyaline casts. There were clumps of lymphocytes throughout the cortices. The arterioles showed some thickening and were occasionally obliterated. The authors interpreted this case as a marked chronic nephrosis. They considered the atrophy and degeneration of the tubules as primary, yet some of the tubules were obstructed by casts and it seems possible that the authors did not give sufficient consideration to this cause of tubular atrophy.

Buschke in 1932 reported a case of multiple myeloma with some renal insufficiency. Albumin in the urine varied from 0.3 per cent to 1.2 per cent. No test was made for Bence-Jones protein. The last concentration test gave a maximum specific gravity of 1020. The blood pressure ranged from 140/70 to 100/60. Two blood urea determinations gave 98 mg. and 62.7 mg. Hemoglobin 27 per cent; erythrocytes 1,100,000. There was no edema. The kidneys were large. Microscopically all the tubules were filled with albumin and casts. The glomeruli were practically normal. There was arteriosclerosis of the large vessels, but the arterioles were normal. Many of the casts were calcified. There were foci of lymphoid cells in the interstitial tissues. The author regarded the lesion as a slowly progressing nephrotic insufficiency. He was not satisfied with the explanation of tubular obstruction by casts.

Mainzer in 1932 described a case of multiple myeloma with marked renal changes. Male, 46 years of age; blood pressure 140/95 to 165/95; proteinuria, at first all Bence-Jones protein, later some globulin but no serum albumin; blood sugar 1.29 to 2.40 per cent, no sugar in the urine; non-protein nitrogen 46.9 mg. to 89 mg.; blood calcium 11.9 to 12.5 mg.; total serum protein 7.38 gm. — albumin 4.69 gm., globulin 2.21 gm., fibrinogen 0.48 gm. The kidneys weighed 140 gm. and 150 gm. each. Microscopically the cortical tubules were small and separated by connective tissue. Large num-

bers of casts filled the tubules. The author's interpretation was nephrotic contracted kidney.

Ehrich studied the kidneys from Mainzer's case and concluded that the lesion was not nephrosis but obstruction of tubules by casts.

Our case (Case 84) bears a close resemblance to the cases of Thannhauser and Krauss, Perla and Hutner, and Mainzer. The tubular atrophy and renal insufficiency are readily explainable as the result of large numbers of casts. In Case 82 the slight interference with renal function may be similarly explained, but in Case 86 an entirely different process is responsible for renal insufficiency, *i.e.* obstruction of the glomerular capillaries.

It seems highly probable that casts are the chief cause of renal insufficiency resulting from multiple myeloma. Completely obstructed tubules undergo dilatation and atrophy. The atrophy of the tubules is not a primary degeneration but a secondary atrophy from disuse. There is no convincing observation on record of a "nephrotic contracted kidney."

2. In the group in which functional tests showed no renal insufficiency there are no satisfactory postmortem reports in the literature. In our case (Case 81) there was some arteriosclerotic atrophy but no change attributable to myeloma.

3. In 25 of the cases collected in Table I there were no functional studies but the kidneys were examined histologically. Löhlein found the tubules obstructed by crystallized Bence-Jones protein. Scheele and Herxheimer, Beck and McCleary, Ellermann and Kleine, all noted the presence of many casts in the tubules. Jochmann and Schumm, and Glaus, found amyloid disease of the kidneys. Two authors found pyelonephritis. This lesion is particularly apt to develop when there is a paraplegia caused by pressure on the spinal cord, and it may also result from prostatic hypertrophy, since the patients are usually elderly men. Arteriosclerosis is mentioned by several authors as a complicating lesion. The diagnoses of "chronic nephritis," "chronic interstitial nephritis," "chronic diffuse nephritis," and so on, which were made by several authors, cannot be accurately interpreted. Probably these terms were applied to changes mainly of arteriosclerotic nature. Scheele and Herxheimer, and Austin, described sclerosis of the superficial zone of the cortex. This change was also observed in our case (Case 81). It is a typical senile lesion resulting from arteriosclerosis.

In summarizing the structural changes in the kidneys found in association with multiple myelomas, it may be stated that some form of renal lesion has been described in nearly every instance. The alteration is usually of a minor nature, and is most often merely the effect of arteriosclerosis or pyelonephritis. When renal insufficiency is present it is probably caused chiefly by casts that obstruct the tubules and may bring about tubular atrophy in extreme instances. In one of our cases (Case 86) there was obstruction of the glomerular capillaries, apparently by plasma of high protein content.

The prevailing opinion is that the amount of Bence-Jones protein excreted is independent of the protein intake (Hopkins and Savory, Folin and Denis, Groat and Brewer, Walters, and Mainzer). Folin and Denis observed an increase when the patient was on a high protein diet but they considered it a coincidence. Magnus-Levy, however, believes that the Bence-Jones protein is not exclusively an endogenous product, but partly derived from proteins of the food.

It is generally agreed that a small amount of Bence-Jones protein may be found in the blood serum, although some investigators doubt the accuracy of the technical methods that are employed. Jacobson found a heavy protein precipitate when the serum was inactivated at 56° C for thirty minutes. He estimated the Bence-Jones protein in the serum at 7.86 per cent. Perlzweig, Delrue and Geschickter found only a small amount of Bence-Jones protein in the serum and suggested that Jacobson's protein was mainly euglobulin. Other investigators agree that only a small amount of Bence-Jones protein is present in the serum (Taylor, Miller and Sweet, 0.2 per cent; Magnus-Levy, a small amount; Hewitt, a trace; Mainzer, 0.006 per cent).

Twelve observations on the serum proteins are available (see Tables I and II). In 9 instances the total protein was normal or slightly decreased, and in 3 there was hyperproteinemia. In the case reported by Perlzweig, Delrue and Geschickter the total serum protein on three examinations ranged from 12.32 gm. to 13.84 gm.; albumin from 1.42 gm. to 4.06 gm.; globulin from 10.11 gm. to 9.09 gm.; and fibrinogen from 0.79 to 0.69 gm. Their analyses indicated that euglobulin forms a large part of the increased protein in the blood. Bannick and Greene found the total serum proteins in 1 case 9.35 gm. — albumin 2.5 gm., globulin 6.5 gm.; and in another the total protein was 10.75 gm. — albumin 4.45 gm., globulin 6.09 gm. They also found the excess protein to be euglobulin.

The Bence-Jones protein does not accumulate in the blood but appears to be excreted readily by the kidneys. The thick protein solution that filled the capillaries and caused renal insufficiency in our case (Case 86) was probably chiefly globulin and not Bence-Jones protein.

The Bence-Jones protein is excreted through the glomeruli. Allard and Weber injected a dog intravenously with 200 cc. of urine containing a large amount of Bence-Jones body and obtained an abundant "albumosuria." Decastello thought that a normal kidney holds back the Bence-Jones body. After injection into dogs he did not find the protein in the urine unless the kidneys had been previously injured by chloroform. Taylor, Miller and Sweet found that large quantities (5 gm.) of Bence-Jones protein must be injected intravenously into dogs before any of it appears in the urine. When the kidneys were injured with uranium nitrate no Bence-Jones body was excreted. Either the injured kidney was less permeable than the normal, or the foreign protein was broken up and eliminated as simpler bodies. Krauss was able to produce Bence-Jones proteinuria in rabbits by injecting large quantities of the protein intravenously. He expressed the opinion that the injured kidney is more permeable to the Bence-Jones body.

It appears established from animal experiments that the Bence-Jones protein may be excreted by normal animal kidneys when it is injected intravenously in large amounts. When small quantities are introduced they are disposed of in some other way. There is no satisfactory experimental evidence that the injured kidney is more permeable to this protein.

The evidence from clinical sources is very strong that the Bence-Jones protein may pass through normal kidneys. There are many reports in which the urinary protein consisted entirely of the Bence-Jones body, and it may be safely assumed that only a slight injury is necessary to permit the escape of serum albumin. The argument that nephritis is more frequent in myelomas with Bence-Jones proteinuria than in those without it has no weight since the most trivial lesions have been reported as nephritis.

Does the excretion of the Bence-Jones protein injure the kidneys? Krauss believed that this protein is toxic and that it injures the kidneys in its passage through them, but Hewitt, and others before him, have shown that the Bence-Jones protein may be excreted in

large amounts over long periods without the appearance of serum albumin in the urine. This fact seems convincing evidence that the Bence-Jones protein may be excreted over long periods without causing injury to the glomerular capillaries. The renal insufficiency that develops so frequently in multiple myeloma is not due to direct injury of the capillaries or tubules but to obstruction of the tubules by casts. The protein injures the kidneys in this indirect way, not during the process of excretion but afterwards.

How are we to explain the presence of serum albumin in the urine? Serum albumin is sometimes the only protein present and often it is found in small or large amounts along with the Bence-Jones protein. Frequently it is present for many months in the early stages of the disease before the Bence-Jones protein appears. There must be at least a slight injury of the glomerular capillaries to allow the escape of serum albumin, and in several of our cases there are marked alterations of the capillary basement membrane (Fig. 1) that indicate capillary damage. But since serum albumin may be present without Bence-Jones protein in myelomas, and *vice versa*, it may be inferred that the Bence-Jones protein does not produce the injury responsible for serum albuminuria. It is not uncommon to find serum albuminuria in malignant diseases, especially those associated with severe anemia. The capillary injury may be caused by anoxemia or by some toxic substance.

The casts in the renal tubules in myelomas are unusual in that they become permanently lodged and act as foreign bodies. Löhlein called attention to casts that were partly crystallized and to some extent surrounded by giant cells. Since no other protein was present in the urine he concluded that the casts were composed of Bence-Jones protein that was partly in crystalline form. Kleine also noted giant cells in contact with large casts. Ehrich observed transitions between granular protein precipitate and solid casts. He agreed with Löhlein that the casts are composed of Bence-Jones protein since there was no other protein in the urine in his case. Giant cells were noted about the casts.

Casts were found in 7 of our 11 cases, and in 1 they were present in enormous numbers (Case 84). In 3 instances some of the casts were partly surrounded by giant cells. The giant cells are apparently formed by the fusion of macrophages that enter the tubule from without. No casts were found in the 2 cases in which no Bence-

Jones protein was excreted (Cases 80 and 81). In Case 89 there were no casts.

There is evidently a causal relation between multiple myeloma and amyloid disease. Magnus-Levy in 1931 collected 19 cases from the literature in which myeloma and amyloid were associated. The amyloidosis may be generalized (Glaus, Weber), but more often it is found in unusual situations such as intestine, voluntary muscle and bones (Askanazy, Hueter, Freund). In Jochmann and Schumm's case and Glaus' case amyloid was found in the kidneys, but usually the spleen, liver and kidneys are not involved. In our case (Case 84) there were extensive deposits of amyloid in the small intestine that caused intestinal obstruction. Magnus-Levy suggested that the Bence-Jones protein is related chemically to amyloid, but all that has been established is that both substances are proteins of endogenous origin.

Summary

1. Renal insufficiency develops frequently in multiple myeloma, especially in advanced stages of the disease.

2. In some instances renal insufficiency is due to arteriosclerosis, in others it is caused by pyelonephritis resulting from compression of the spinal cord, or from prostatic hypertrophy.

3. The only direct effect of multiple myeloma on the kidneys is due to the formation of tubular casts of Bence-Jones protein that obstruct the tubules and cause tubular atrophy. When large numbers of tubules are obstructed extensive atrophy of the cortex and renal insufficiency ensue.

4. There is no evidence that the Bence-Jones protein injures either tubules or glomeruli. Cortical atrophy is on an obstructive basis and should not be interpreted as "nephrotic contracted kidney."

5. In one instance renal insufficiency was apparently caused by the accumulation of a highly concentrated protein solution in the glomerular capillaries. In two other instances this same appearance was found in a few capillaries.

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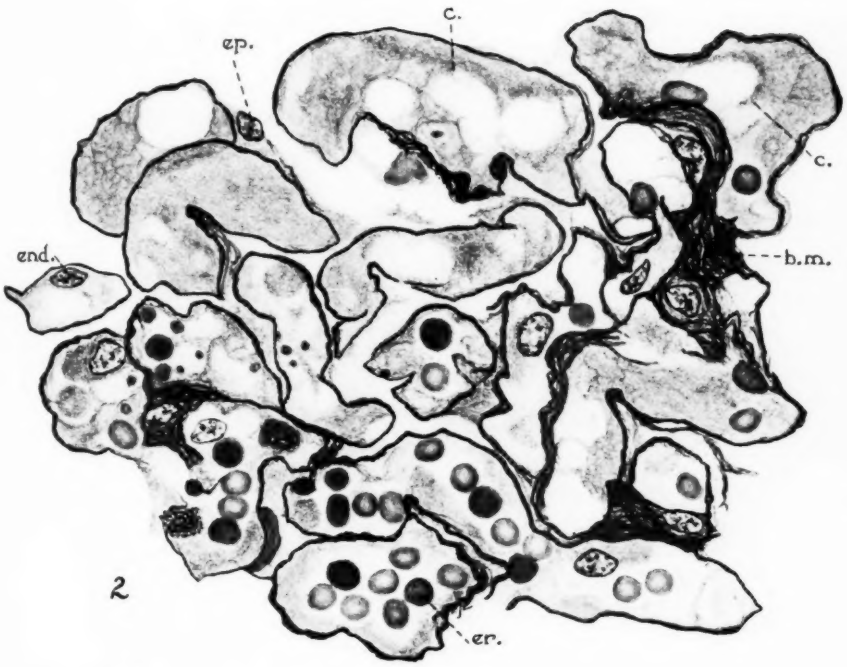
DESCRIPTION OF PLATES

PLATE 63

FIG. 1. A. 32–930. Portion of a glomerulus showing an irregular thickening of the capillary basement membrane: b. m. = normal basement membrane; b. m′. = thickened basement membrane; end. = endothelial nucleus; ep. = epithelial cells; er = erythrocytes.

FIG. 2. A. 32–1024. Portion of a glomerulus showing all the capillaries distended with protein; c = protein coagulated by the fixative. Other lettering as in Fig. 1.

FIG. 3. A. 29–1165. Glomerular capillaries filled with coagulated protein; c, as in Fig. 2. Lettering as in Fig. 1.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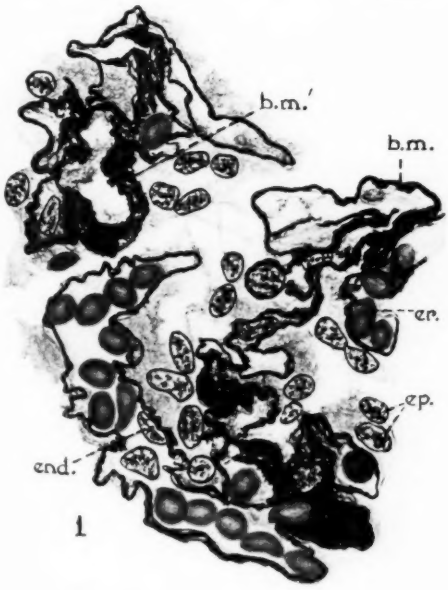
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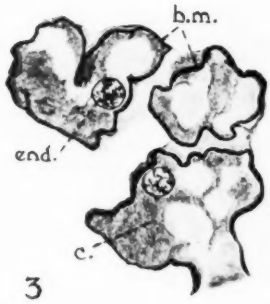
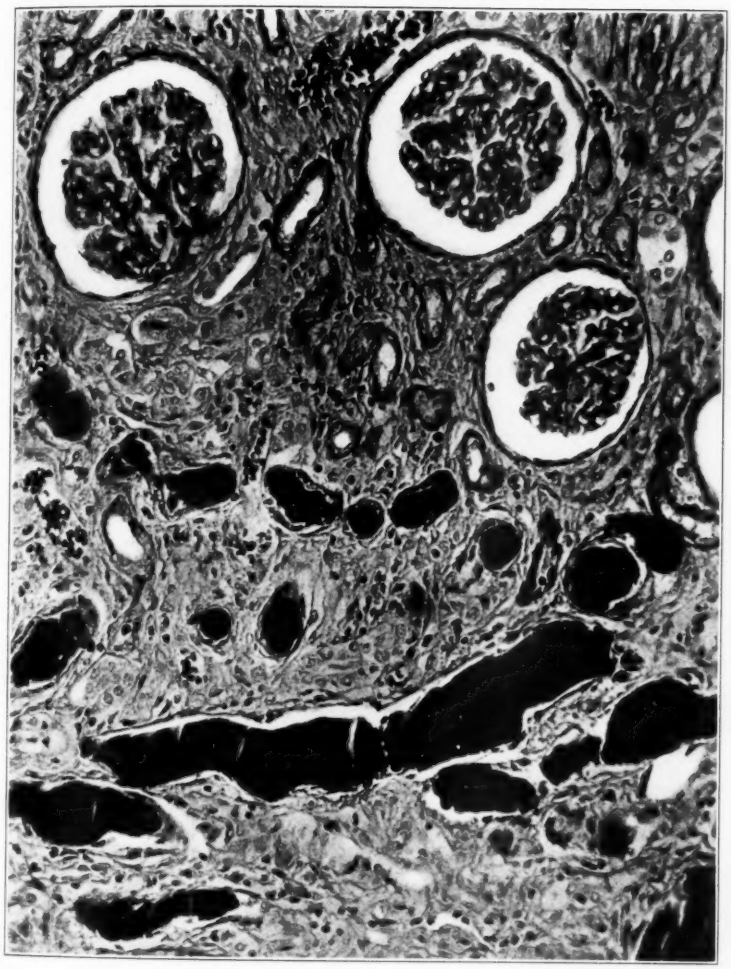

b.m.
end.
c.
3

PLATE 64

FIG. 4. A. 31–408. Area of cortex of kidney showing tubules filled with casts, and atrophy of the tubules proximal to the casts. The glomeruli show only a moderate thickening of the capillary basement membranes.



4

NEUROBLASTOMA METASTASES IN BONES, WITH A CRITICISM OF EWING'S ENDOTHELIOMA *

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INTRODUCTION

The characteristics of Ewing's sarcoma or diffuse endothelioma of bone, as described by Ewing [1] and other writers, are as follows. The patient is usually a child. The tumour appears most frequently in a short or long bone of a limb and, if in a long bone, affects the shaft diffusely. The onset is with pain and disability, and later enlargement of the segment of the limb. Intermittent mild pyrexia is often present. These features lead in many cases to an initial diagnosis of osteomyelitis, a diagnosis that skiagrams, unless interpreted by workers familiar with this class of tumours, may be held to confirm. Exploratory operation reveals a soft tumour mass surrounding the bone and often infiltrating surrounding soft tissues. Histological examination of an excised specimen reveals a richly cellular tumour composed of closely aggregated, small spheroidal cells of uniform size and shape, each with a spherical hyperchromatic nucleus. These cells appear in diffuse sheets or masses usually devoid of any specific structural arrangement. Purely histological diagnosis, therefore, is not possible. The tumour is highly susceptible to adequate doses of X-radiation, which produces prompt diminution or disappearance of the growth. This favourable response is held to be almost diagnostic of Ewing's tumour. Recurrence, however, is the rule and death usually occurs within two or three years. Almost invariably secondary growths, regarded by some as metastases and by others as multiple new formations, appear in many other bones, especially in the skull and, unlike other bone tumours, Ewing's tumour frequently yields metastases in lymph glands.

The object of this paper is to report a tumour that presented all

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the accepted features of Ewing's tumour, but which careful autopsy study revealed to be one of many skeletal metastases from a small adrenal neuroblastoma.

Report of Case

Clinical History: J. T., a well developed girl 8 years old, had had previous good health and had suffered no noteworthy illnesses. In June 1932 persistent pain in the right thigh began and in August a swelling of the upper part of the thigh was noticed. The child was admitted to the hospital with the diagnosis "chronic osteomyelitis." Save for a slightly tender, diffuse enlargement of the upper half of the right thigh, general examination disclosed no other abnormalities. The skiagraphic report was: "Area of bone destruction and periosteal reaction in upper third of shaft of femur, appearance suggestive of osteomyelitis. A leukocyte count gave 10,400 cells per cmm. The Wassermann test yielded no reaction. Slight pyrexia was present, the evening temperature frequently reaching 99° F, and sometimes 100° F. On Sept. 17, 1932 the skiagraphic report was as follows: "There has been some extension of the bone involvement, the upper two-thirds of the shaft now being affected." (See Fig. 1.)

On September 21 exploratory operation was performed, revealing a large soft tumour enveloping the shaft of the femur and invading the surrounding soft tissues. A fragment was excised for histological diagnosis. The report on this specimen was: "Richly cellular, round-celled, highly malignant tumour invading skeletal muscle; specific nature cannot be affirmed, possibly a soft tissue sarcoma, possibly a metastatic growth."

A skiagram of the thorax on October 5 disclosed no evidence of lung metastases. On the same day a course of deep X-radiation to the tumour was commenced, and was continued until Nov. 21, 1932. Within four days of the commencement of this treatment pronounced reduction in the size of the growth was apparent and pain and tenderness were much relieved. The rapidity of diminution of the tumour following the initial applications of X-rays was dramatic. During this course of treatment, and thereafter until her death, the patient had increased pyrexia up to 101° F in the evenings, and sometimes to 102° and 103°.

In skiagrams taken on Oct. 27, 1932 no pulmonary metastases were visible but a paravertebral tumour was noted (Fig. 2). The subsequent course was one of emaciation, cachexia and further increase in the size of the femoral tumour. Death occurred on Nov. 29, 1932. The clinical diagnosis was "Ewing's sarcoma of the femur."

Postmortem Examination

Autopsy was performed six hours after death. There was an enormous, fusiform, soft white growth surrounding the whole of the shaft and neck of the right femur and invading the surrounding muscles. The tumour tissue was easily detached from the femur, the exposed surface of which was eroded and roughened and resembled coarse sandpaper to the touch (Fig. 3). At the lower limit of the

growth there was a slight degree of periosteal new bone formation. The medullary cavity was occupied by tumour tissue. All other bones of the skeleton that were examined presented similar but less conspicuous tumour deposits, situated chiefly beneath the intact periosteum. The inner and outer surfaces of nearly all the cranial bones, mandible, all vertebrae, all ribs, sternum, right humerus, clavicles, pelvis and left femur were involved. Only on the lumbar and thoracic vertebrae did the subperiosteal growths produce tumour masses readily visible externally; elsewhere they formed a thin stratum only discovered on incising the periosteum. The vertebrae and several ribs were sectioned and were found to contain medullary deposits of tumour also. The peripheral bones of the limbs were not examined.

Numerous discrete white tumour nodules were present in the lungs and liver. Both kidneys contained a few tiny points of growth. There were small metastases in the iliac and lumbar lymph glands. The medulla of the left adrenal contained two or three tiny discrete white nodules. The right adrenal was the seat of a group of white nodules occupying the medulla; these, though they appeared to be separate when viewed on a single cut surface (Fig. 4), were connected for the most part with one another, forming a lobulated tumour 3 cm. in maximum diameter with a few smaller, separate satellite nodules. All other organs, including the skull contents, appeared normal.

Histological Examination

Microscopic study of haematoxylin-eosin sections of all of the tumours described above, as well as of various viscera, was carried out. Frozen sections of the adrenal growth were stained also by Bielschowsky's method and by Cajal's silver pyridine method for nerve fibres.

The growths in all situations consisted of diffuse masses of rounded cells, each 10 or 12 microns in diameter, with a spherical, deeply stained but vesicular nucleus that sometimes presented a single small nucleolus. Mitotic figures were numerous. The bulk of the tumours exhibited no special arrangement of the cells, but in parts of the right adrenal growth distinct rosette formation was present (Fig. 5). The same feature was observed also, though less clearly, in some of the hepatic metastases. No nerve fibres were found in the Bielschowsky and Cajal-stained sections, and there was no evidence

of any tendency of the tumour cells to differentiate toward the adult nerve cell type.

Of the viscera that appeared macroscopically normal, only the spleen exhibited abnormalities. The vascular spaces of the splenic pulp contained sparsely scattered, small clumps of tumour cells, which were found also in a small accessory spleen.

Discussion

The identity of the tumour in this case is scarcely open to doubt. The primary growth was a highly malignant but small neuroblastoma of the right adrenal that had produced bulky metastases in the other viscera, and especially in the skeleton. The cytology and rosette formation of the adrenal tumour are characteristic of the neuroblastomas of childhood. That the adrenal tumour was small is no argument against its primary character, for the primary growths in the Hutchison [2] and Pepper [3] types of adrenal neuroblastoma are notorious for their small size, compared with that of their metastases in the skull or liver. The possible suggestion that because the left adrenal contained metastatic nodules the right adrenal tumour was also metastatic in nature does not accord well with the much larger size of the right-sided tumour. If, in spite of the cytology of the adrenal growth, it is assumed that the femoral tumour was primary, it is still necessary to admit that all the other skeletal growths were secondary, and this admission greatly detracts from the argument for the primary nature of the femoral tumour. That this tumour was much larger than any other of the skeletal deposits, and that it was the clinically predominant tumour, provide no argument for its primary character, for the Hutchison skull tumours also are in the forefront of the clinical picture and are much larger than the coexistent deposits in other parts of the skeleton.

As regards this clinical predominance of one out of many metastases in bone, a possible cause may be found in the relations of tumour to periosteum in the various situations. It may be that the reason why one metastasis grew luxuriantly while the others remained clinically dormant is that for some cause, possibly traumatic, the periosteum had undergone a solution of continuity at the site of the dominant growth, thereby releasing it from a restraining influence and allowing the neoplasm to flourish unchecked in the

surrounding tissues. In other words, the tumour developed precociously in one particular situation because the restraining periosteum suffered penetration. However, we can see no escape from the conclusion that the case described was one of adrenal neuroblastoma with prolific skeletal metastases, one of which outstripped the others and simulated a large primary tumour of the femur.

Clinically the femoral tumour was diagnosed as a Ewing's sarcoma. How valid was this diagnosis? The accepted characteristics of Ewing's tumour have been summarised in our introduction, and our case presented all of these characteristics. The patient was a child, the tumour affected diffusely a large part of the shaft of a long bone, the onset was with pain and disability, the course of the disease was febrile, the initial clinical and skiagraphic diagnosis was osteomyelitis, metastases developed in other parts of the skeleton including the skull, pulmonary metastases were late in their appearance, biopsy revealed a diffuse spheroidal-celled growth with small spherical hyperchromatic nuclei, and the initial response to X-ray therapy was prompt and striking. In all these respects our tumour conforms to Ewing's description, and further, the skiagraphic features of our case and also the gross appearance of the eroded bone shaft closely resemble those depicted in Ewing's Figures 127 to 129.[1] We believe then that, on all possible clinical grounds, the diagnosis of Ewing's tumour was fully justified, and this diagnosis might easily have received false corroboration from the autopsy, had this not been complete.

It is pertinent then to inquire whether other alleged instances of Ewing's sarcoma or endothelioma may not also have been of a nature similar to ours, and whether adequate autopsy study has been made in any of the cases that have received this clinical designation. In pursuing this inquiry it is disappointing to find that neither in Ewing's accounts nor in those of Connor,[4] Coley and Coley,[5] Kolodny,[6] and Clopton and Womack [7] is there any detailed autopsy record of any of their cases, and that in the majority of instances the diagnosis rested on clinical and biopsy findings only. Thus, Ewing [8] records no details of autopsy findings. Connor, discussing 52 bone tumours classified as Ewing's sarcoma in the Registry of Bone Sarcoma, states that "necropsies were done in less than one-third of the cases," and his paper gives no indication as to how complete these autopsies were. Coley and Coley describe many

cases of "endothelial myeloma" diagnosed on clinical and biopsy findings, but autopsy is recorded in only one case (No. 49), of which it is stated: "A necropsy was performed, revealing very extensive metastases in nearly every bone and organ." Kolodny gives a full account of clinical and skiagraphic findings on which, along with the radiotherapeutic response, he would rely entirely for the diagnosis. He considers that clinical distinction from metastatic growths in bones can be made because "metastases occur in the very young or after the age of 40, while most Ewing's sarcomata are seen in early adolescence," a statement with which we venture to think very few pathologists will agree. No autopsies were performed on Clopton and Womack's cases. In the only postmortem record specifically mentioned by Ewing in his Neoplastic Diseases (page 361), it is perhaps noteworthy that "retroperitoneal lymphatic metastases were found." On page 352 Ewing says "the diagnosis of endothelioma of bone should not be made until a thorough search for a primary tumor has proved unsuccessful, and this search may, at times, not be regarded as complete without autopsy." We concur with this view, but would modify the latter half of Ewing's statement to read, "this search may *never* be regarded as complete without *thorough* autopsy."

Since no adequate records of autopsies of cases classed as Ewing's tumour are available, we may turn to earlier literature for possible information on the subject. Of much interest is a paper by Roman [9] who described under the title "myeloplastic sarcoma" two cases of widespread round-celled growths of many bones in children. In one case there was a supposedly metastatic growth involving the left adrenal, and in the other a tumour completely replacing the adrenal. Roman reviewed also several recorded tumours that resembled his own, those of Gussenbauer, Dittrich, and Schmidt, in all of which it is of interest to note that tumour nodules were present in the adrenals. Indeed, it is clear that the tumours described and reviewed by Roman were not, as he supposed, "myeloplastic sarcomas," but instances of adrenal neuroblastoma with widespread bone metastases in the same category as the Hutchison tumours. Evidently Roman was not aware of Hutchison's work of five years previously.[2] Yet, strange to say, Kaufmann [10] accepts Roman's interpretation.

A purely histological diagnosis of Ewing's tumour is not possible.

All writers on the subject admit that there is nothing distinctive about the cytology or architecture of the growths. As MacGuire and McWhorter [11] say: "Most pathologists would call these tumors round-celled sarcomas." However, of possible significance (as regards a neuroblastic nature) is Ewing's [1] observation (see page 359) that "rosette structures without lumina" are present in some of the tumours.

MacGuire and McWhorter confess themselves perplexed by the scope and identity of the group of tumours under discussion, and they conclude: "Ewing's tumor is not yet established as a clinical entity." Hirsch and Ryerson [12] also severely criticise the records of alleged cases, and they suspect that most of them were instances of metastases from undiscovered primary tumours. With this criticism we agree and, while we think it probable that a variety of different conditions has been included in the group of Ewing's tumours, we would urge strongly the claim of adrenal neuroblastoma as the responsible tumour in many instances. This claim is supported by the general characteristics of cytology and metastatic distribution of Ewing's tumours. The multiplicity of skeletal growths, their diffuseness and often predominantly subperiosteal situation, the frequent involvement of the skull, and the frequent presence of lymph gland metastases (a rare feature with all primary bone tumours), are all reminiscent of the malignant neuroblastomas as described by Hutchison,[2] Tileston and Wolbach,[13] Frew,[14] Wollstein,[15] and others. It may be recalled that Hutchison's tumours were formerly called "sarcomas" of the skull. We believe that future careful autopsy work will furnish a revelation regarding the Ewing sarcomas similar to that furnished by Hutchison's work for the skull tumours. If, in a case of the Hutchison type, we imagine the bulky skull tumours to be transferred to a long bone, a Ewing tumour would result. The age incidence of the two classes of growth is slightly different, Hutchison's tumours appearing usually in infants, and Ewing's tumours chiefly in older children. It is possible, however, that this very difference may be related in some way to the different sites of preference of the apparent "primary" tumours.

We would draw attention also to the significant fact that "Ewing's sarcoma, primary in the skull" has rarely if ever been recorded, although all writers on the subject comment on the remarkable frequency with which secondary growths appear in the skull. Perhaps

the explanation of this seeming anomaly is that when skull growths are the first to appear the case is recognised correctly as belonging to the Hutchison group and thereby escapes the designation "Ewing's sarcoma."

Finally, we have searched in vain for observations other than our own on the radiosensitivity of neuroblastic tumours, but, from the truly embryonal qualities of their cells and from their high mitotic activity, we would not be surprised to learn that they frequently possess the susceptibility to X-radiation that has been regarded as almost diagnostic of the Ewing tumour.

Summary and Conclusions

1. A case is described in which a tumour presenting all the accepted characteristics of a Ewing's sarcoma of bone was shown at autopsy to be one of many metastases from an adrenal neuroblastoma.

2. Review of certain adequately recorded autopsy cases of supposed multiple bone sarcomas leads to the conclusion that these also were instances of adrenal neuroblastoma with skeletal metastases.

3. The term "Ewing's sarcoma," while possessing clinical value as defining a syndrome presented by a certain group of tumours affecting bones, has no established claim as designating a pathological entity.

4. While not denying the *possible* existence of a primary bone tumour presenting the Ewing syndrome, we believe that further study will disclose the metastatic nature of most of the tumours with this syndrome, and we strongly suspect that adrenal neuroblastomas will prove to be the primary growths in many of the cases.

We are indebted to Dr. L. Love for his interest in the skiagraphic aspect of the case reported and for preparing prints of the skiagrams shown in Figures 1 and 2.

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DESCRIPTION OF PLATE

PLATE 65

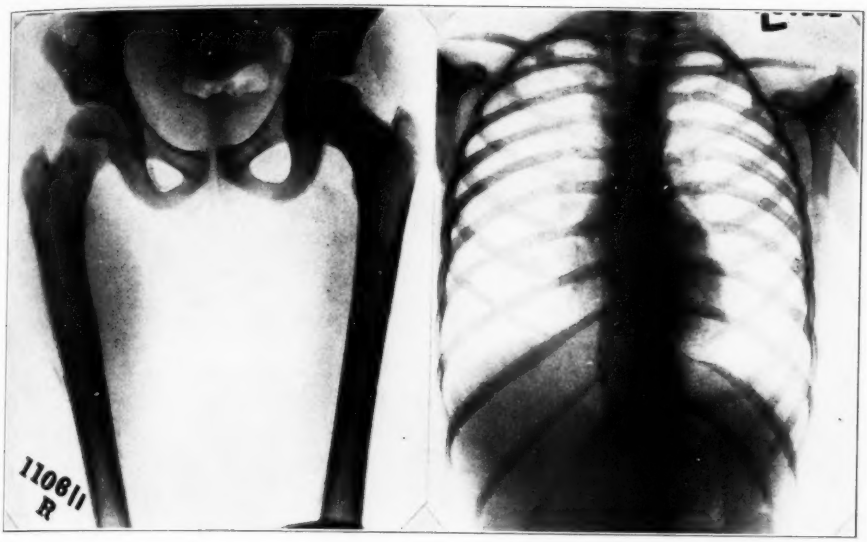
FIG. 1. Skiagram of femur on Sept. 17, 1932.

FIG. 2. Skiagram of thorax on Oct. 27, 1932, showing paravertebral tumour.

FIG. 3. Anterior view of femur after removal of soft tumour tissue. Half natural size.

FIG. 4. Vertical section of right adrenal. Natural size.

FIG. 5. Two views of haematoxylin-eosin stained sections of adrenal tumour showing rosettes.



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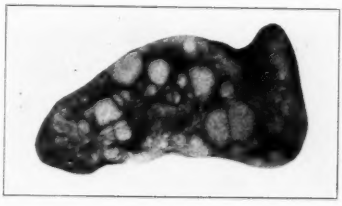

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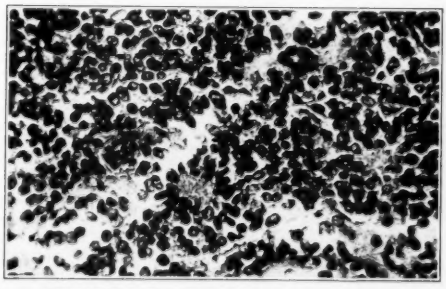

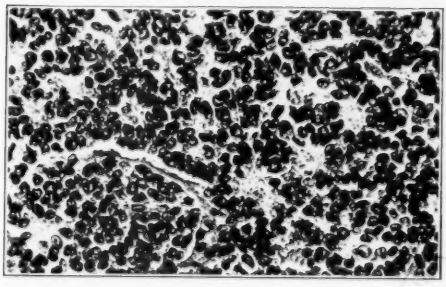

5

Colville and Willis

Neuroblastoma Metastases in Bones

EXTREME ALTERATION OF THE AORTIC VALVE IN SYPHILITIC AORTITIS *

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A characteristic gross picture of changes of the aortic valve in some cases of syphilis was described in a previous communication.[1] It was shown that adhesions occur between the lateral portions of the aortic cusps and the corresponding intima of the sinus of Valsalva. These adhesions lead to a spreading of the commissures of the aortic valve and are considered pathognomonic of syphilis. Two cases showing extreme degrees of separation of the commissures, obscurity of a sinus of Valsalva in one instance and transformation of one sinus into a cavity that was filled with blood in the other instance, form the basis of the present communication.

CASE REPORTS

CASE 1. The patient was a 43 year old male. There was clinical evidence of aortic insufficiency over a period of 10 months. The blood Wassermann and Kahn reactions were positive (4 plus).

At autopsy there was edema of the lower extremities, ascites, hydrothorax, and chronic passive hyperemia of the viscera. The aortic changes were characteristic of syphilis. These changes were most pronounced in a segment about 4 cm. in length just above the aortic valve. The mouths of both coronary arteries were markedly narrowed.

The heart weighed 550 gm. The mural endocardium of the interventricular septum in the left ventricle was thickened and showed several endocardial pockets that were open toward the aortic valve. There were occasional yellow plaques of sclerosis on the mitral valve leaflets, but no changes of the pulmonary and tricuspid valves.

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When the aortic valve was opened only two cusps and their corresponding sinus of Valsalva were visible. The posterior cusp on first inspection could not be distinguished and seemed absent, suggesting the possibility of a congenital anomaly. A closer inspection, however, revealed a small, slit-like opening about halfway between the right and left cusps. The distance between the right and left cusps measured 33 mm., while the slit-like opening measured only 6 mm. across and led into a very narrow sinus. The anterior wall of this sinus, therefore, was interpreted as being the remnant of the posterior cusp. The commissure between the left and right cusps was wider than normal, measuring 5 mm. in diameter. The free margins of these cusps were thickened and slightly everted. The commissures were elevated, rough, thickened and partially hyalinized. The aorta just above the commissures revealed many areas of hyalinization, fatty changes, some calcification, and many longitudinal depressed scars and wrinkles with reddish bases. The mouths of both coronary arteries seemed displaced and were much narrower than normal. Both coronary arteries revealed only a moderate degree of arteriosclerosis. The papillary muscles and columnae carneae of both ventricles were hypertrophic and flattened. There were many gray streaks of fibrosis throughout the myocardium.

The area of the aortic ring with the aortic valve was cut into four blocks. All the blocks contained the aortic wall of the sinus of Valsalva and a considerable portion of the adjacent aorta, in addition to the following structures: Block I contained portions of the left and right cusps and the commissure between these cusps. Block II contained the midportion of the right cusp. In Block III a small remaining part of the right cusp, the very large commissure between the right and posterior cusps and also the remnants of the posterior cusp were embedded. Block IV consisted of the commissure between the posterior and left cusps and also of a small portion of the left cusp.

Serial sections of each of the four blocks were made. The sections were stained alternately with hematoyxlin-eosin, with a combination of orcein and iron hematoxylin, and according to the Van Gieson method.

Histological Examination of the Aortic Valve

Block I (Portion of Left Cusp, Aorta, Commissure and Portion of Right Cusp): The intima of the aorta revealed a marked thickening with fibrosis and hyalinization. Between the intima and the media many newly formed capillaries were present, many of which were dilated and surrounded by lymphocytes and a few plasma cells. The media showed an interruption of the continuity of the elastic lamellae. Small vessels surrounded by lymphocytes were found in these regions. Serial sections revealed that many of the small vessels crossed the media transversely. The adventitia also showed small sized vessels, the lumina of which were partially or completely obliterated. There was a marked perivascular infiltration of round cells and a few plasma cells, more pronounced than in the other layers, and also a new formation of connective tissue with some hyalinization. Corresponding to the region of the base of the left cusp, small sized blood vessels, many lymphocytes and plasma cells and much connective tissue were found in the intima and media of the aorta extending into the base of the cusp. The intima and media close to the commissure revealed areas characteristic of granulation tissue, namely, many small vessels perpendicular to the surface, that were surrounded by lymphocytes and plasma cells.

Sections that were taken through the lateral portions of the left cusp revealed areas consisting of lymphocytic infiltration and many newly formed capillaries. A few longitudinally cut vessels were found, apparently extending along the lateral portion of the cusp toward the commissure. Close to the commissure much fibrosis and hyalinization were encountered and only relatively few cellular elements. Small blood vessels were still recognizable in these regions. Within the commissure the proliferative changes were much more marked than in other portions of the aorta and in the lateral part of the cusp. The intima showed large areas of hyalinization with calcification and also a new formation of irregularly shaped elastic fibers. Very few small vessels were present in this region, and only occasionally a few lymphocytes could be recognized. The adventitia in these regions showed much fibrosis and hyalinization. Many vasa vasorum, both arteries and veins, were obliterated. Just above the commissure small blood vessels were present, some cut transversely

and some longitudinally, the latter apparently extending from the intima into the adjacent cusp.

Sections taken through the regions of the attached cusp revealed a microscopic opening between the intima and the aortic cusp in areas of the widened commissure in which, grossly, the sinus of Valsalva was not visible. Much hyalinization and fibrosis, but only occasional small blood vessels, were found within the adherent portions of the cusp. Relatively more vessels were found in the distal and proximal portions of the adherent cusp than in its midportion. In only a few sections were a slightly larger number of vessels present at the base of the cusp, these apparently extending from the aorta into the cusp. The lateral portion of the remnants of the right cusp, which was still within the block, revealed at the base loose connective tissue with dilated capillaries and a slight perivascular infiltration.

Block II (*Midportion of Right Cusp and Corresponding Aorta*): The upper portion of the cusp revealed marked fibrosis and hyalinization, in addition to slight infiltration of lymphocytes and a few endothelial cells. Some of the latter cells were arranged in the form of whorls. The base of the cusp showed dilated capillaries, while the central portion of the cusp revealed a slight increase in connective tissue, but no cellular infiltrations of note. The changes in the aorta were similar to those described before. The region of the orifice of the right coronary artery showed a marked new formation of connective tissue with hyalinization and some calcification. The first portion of the coronary artery very close to its mouth revealed a bulbous-like dilatation.

Block III (*Lateral Portion of Right Cusp, Commissure between the Right and Posterior Cusps, Posterior Cusp and Aorta*): Some portions of the right cusp revealed capillaries, some of which were surrounded by lymphocytes. Sections that were taken from the commissure itself showed only occasional capillaries extending from the aortic wall into this region and infiltrations of lymphocytes. The central area of the cusp revealed much fibrosis and hyalinization and also a few vessels that extended partly from the proximal and partly from the distal portion of the cusp into the central region. Many of the sections contained a slit-like opening between the cusp and the intima of the sinus of Valsalva, while others showed a new formation of connective tissue with small vessels between the aortic wall of the sinus of Valsalva and the aortic cusp in more localized areas sub-

dividing the space of Valsalva into small compartments. In the sections that were taken from the lateral borders of the cusp a differentiation could not be made microscopically between the thick cusp and the thickened intima of the wall of the sinus of Valsalva. Some of these regions revealed only hyalinization and fibrosis without blood vessels or cellular infiltration. The aortic changes were similar to those described before. The free margin of the posterior cusp revealed slight fibrosis, but few cellular elements.

Block IV (*Commissure between the Posterior and Left Cusps, and Portion of Left Cusp and Aorta*): Within the commissure blood vessels extended from the intima of the aorta into the adjacent portions of the cusp. Some of them were surrounded by lymphocytes, plasma cells, a few endothelial cells and occasional polymorphonuclear leukocytes. These changes were more marked the closer the sections were taken to the adjacent cusps. A typical granulation tissue was found in this region. Sections that were taken from the base of the cusp also revealed blood vessels and a large number of lymphocytes and endothelial cells. It was demonstrated in the serial sections that some of the blood vessels extended from the aortic wall, at the base of the sinus of Valsalva, into the adjacent basic portion of the cusp. Also, the cellular infiltrations were continuous with those of the aortic wall. The aortic changes were similar to those described before. In several sections, very close to the lateral part of the cusp, the aortic intima was swollen and contained spindle and stellate cells that were separated from one another by an edematous material stained slightly pink with eosin.

CASE 2. This was a colored male, 44 years old.* The clinical diagnosis was syphilitic aortitis with insufficiency of the aortic valve.

At autopsy a typical syphilitic aortitis and a marked narrowing of the mouths of both coronary arteries were found. The commissures of the aortic cusp were much wider than normal. The sinus of Valsalva corresponding to the posterior aortic cusp was almost completely transformed into a cavity by adhesions between the free margin of the cusp and the corresponding intima of the aorta. Only a small midportion of the cusp was free from adhesions. The cavity thus formed was distended because of a small amount of clotted

* This patient was admitted to the Cleveland City Hospital. I am indebted to Prof. H. T. Karsner for permission to publish this case.

blood. Sections were taken from various portions of the aortic valve and adjacent aorta, and stained similarly to those of Case 1.

Histological Findings: The histological examination revealed changes practically identical with those seen in the preceding case. The adventitia and media revealed changes typical of syphilitic aortitis. In the aortic intima there were marked proliferative changes, fibrosis and hyalinization. Small blood vessels were recognized that extended into the commissures. Some of them were surrounded by round cells. Within the widened commissure much old connective tissue was found with slight lymphocytic infiltration. In the region where the cusp comes off the widened commissures the cellular infiltration was more pronounced and a few endothelial and plasma cells were noted. The central portion of the non-adherent cusps revealed no noteworthy changes, while the upper free margins were the seat of simple hyalinization and fibrosis. The base of the cusps showed a few blood vessels, some of which were surrounded by lymphocytes. In the region where only the upper margin of the cusp was adherent to the aorta areas suggesting granulation tissue were found with many dilated blood vessels, lymphocytes, endothelial cells, plasma cells and also a few polymorphonuclear leukocytes.

Discussion

The two cases are similar in many respects. Both revealed a typical syphilitic aortitis grossly and histologically. Both showed a widening of the commissures. In Case 1 the posterior cusp was almost completely attached to the aortic wall of the sinus of Valsalva, leaving only a relatively small midportion free from adhesions. In Case 2 the adhesions were confined to the lateral upper margins of the posterior cusp and did not affect the central portion of the cusp. These changes resulted in a transformation of the sinus of Valsalva into a cavity. The blood clot found within this cavity revealed no evidence of organization. *Intra vitam*, blood must have collected there, which could not be expelled. The blood that filled the cavity evidently prevented the cusp in its entire extent from becoming adherent to the aortic wall, as in the first instance.

The outstanding histological features in both cases are alike. The syphilitic changes in the aorta are the perivascular round cell infiltrations of the adventitia and media, endarteritis obliterans of

the vasa vasorum, and repeated interruption of the continuity of the elastic lamellae of the media.

As compared with the early changes of syphilitic aortitis, which we had occasion to examine in the study referred to previously,[1] the endarteritic changes in general were less frequently encountered in these two cases, whereas the fibrosis of the adventitia was much more marked. This discrepancy of findings in early and in old cases may explain the divergence of opinion as to the primary site of syphilis of the aorta. While Backhaus,[2] Waite,[3] and others believed that obliterative endarteritis of the vasa vasorum is the primary lesion in the aorta, other investigators, and very recently Scherer,[4] hold the media as the primary site of syphilis. Scherer stated that the primary changes of the media surely are the direct result of the spirochetes, and the damage caused by spirochetes is followed by inflammatory processes. The author, however, failed to demonstrate the spirochetes. Furthermore, his material was taken from patients afflicted with generalized paresis and, therefore, cannot be considered as early syphilis.

The intima, though not revealing lesions morphologically characteristic of syphilis, showed proliferative changes, fibrosis and hyalinization, occasional calcification and granulation tissue. The extension of the granulation tissue into the adjacent and adherent portions of the cusps could be made out clearly. In the central parts and also in the region of the free margins of the cusps, the granulation tissue was absent. In other words, the lateral portions of the aortic cusps were the seat of a chronic inflammation with new formation of vessels. These vessels not only reached the cusps by means of the commissures but also extended from the aorta into the basal portions of the cusps. The extension of blood vessels and groups of small round cells into the base of the aortic cusp was noted by Longcope [5] as early as 1910. In a recent communication Benedict [6] classifies two types of inflammatory processes in the aortic valve, namely, an ascending type in which the process spreads through the base of the cusp, and a descending type characterized by the extension of the inflammation through the commissures. It should be emphasized that the evidence presented demonstrates that an inflammatory process arose in the aorta leading to the formation of granulation tissue which, extending to the cusps, formed adhesions between the aorta and the cusps. Also, the newly formed connective tissue that ex-

tended from the aortic wall of the sinus of Valsalva into the cusp, subdividing the sinus into smaller compartments in our first case, is significant in this respect because a cord of connective tissue in this region can be explained only on a basis of granulation tissue.

Very little can be said about the histogenesis of the valvular lesions from our two cases because the lesions are old. It must be realized that only from early lesions may deductions be drawn as to pathogenesis. In the investigation that was mentioned [1] 71 cases of syphilitic involvement of the aortic valve were studied. In this group some very early cases were included that revealed nutritional disturbances in the intima and lateral portions of the cusps. The resulting degenerative lesions (mucoid degeneration and necrosis) became organized secondarily and were followed by chronic inflammation. Only occasionally could such degenerative lesions be demonstrated in these two cases, apparently because of the long duration of the disease. Krischner [7] in a recent article considered two possible causative factors — (*a*) a sensitivity of the tissue of the valve to the syphilitic toxin, or (*b*) a primary dilatation of the anulus fibrosus with resulting relative hypertrophy of the valve. The separation, wrinklings and thickenings of the commissures, he thought, were due to the stretching of the valvular areas brought about by the relative insufficiency. In both of our cases the dilatation of the aortic ring area was not sufficiently marked to explain the widening of the commissures simply by stretching of this area. Also, the adhesions of the cusps to the aortic wall, in our opinion, cannot be explained as the result of a primary dilatation of the aortic valve region. The findings of typical granulation within the disfigured portions of the aortic valves in both cases speak against a primary mechanical cause. The possible sensitivity of the valvular tissue to a syphilitic toxin cannot be studied with morphological methods. We do not believe such an explanation should be advanced at the present stage of our knowledge of spirochetes and their possible toxins. In our two cases an extension of the granulation tissue was noted not only into those portions of the cusps and commissures that showed the very severe gross lesions, but also into all the commissures. In addition to much fibrosis and hyalinization, found in many sections, blood vessels accompanied by round cell infiltration were noted. These findings also speak against a mechanical genesis of the widening of the commissures and make an inflammatory origin more likely. Whether or

not the hypothesis that suggests the primary changes in the aortic intima and within the aortic cusps are nutritional disturbances followed by chronic inflammatory changes is correct cannot be deduced from the examination of these two old cases. It must be maintained, however, that a careful histological examination of the commissures that are characteristically widened in syphilis reveals inflammatory changes that cannot be explained on a mechanical basis alone.

In regard to adhesions between the aortic cusps and the intima of the aorta it might be of interest to mention Maresch,[8] who pointed out that during systole the cusp is pressed toward the intima. During diastole, however, the thickened intima above the sinus prevents the regurgitating blood columns from opening the sinus. The cusp therefore remains fixed and later becomes adherent.

Marked deformities of the aortic valve due to syphilis, such as are here described, are very rare. We were able to find only eight similar instances in the literature. Koch [9] in 1911 reported a case of syphilitic aortic insufficiency in which the left aortic cusp was almost completely adherent to the aortic intima. Only a slit-like opening between the cusp and the sinus of Valsalva was found on section. Paltauf [10] in 1913 reported a similar case in which the right cusp was adherent. In Engel's [11] case, also reported in 1913, the sinus of Valsalva was so small as to allow only the introduction of a very thin probe. Maresch in 1930 published four cases, two of which he had observed himself. The other two cases were museum specimens, one dating from the time of Rokitansky. This latter case was, according to Maresch, described as follows: "Cor quinquageniariae dilatatum ac hypertrophicum cum vasorum truncis. In ostio aortae dilatatae ac incrassatae valvula dextra ex concretione cum vasis pariete abolita, ostium arteriae coronariae collateralis obliteratum conspicitur." (A dilated and hypertrophic heart with the large vessels of a 50 year old female. At the root of the dilated and thickened aorta the right valve, because of adhesions to the vessel, appears absent and the mouth of the coronary artery obliterated.) The eighth case was reported by Herxheimer [12] in 1931.

Summary

Two cases of syphilitic aortitis with involvement of the aortic valve are reported. In both instances the involvement of the valve had reached extreme degrees, caused a transformation of one sinus of Valsalva into a cavity in one instance, and an almost complete disappearance of a part of one sinus of Valsalva in the other. Only eight similar cases were found in the literature. Histologically granulation tissue and much fibrosis and hyalinization were found within the commissures and the adjacent parts of the adherent aortic cusps. The granulation tissue had spread from the aortic intima through the commissures and also through the base of the cusps into the cusps themselves. The widening of the commissures, which is pathognomonic of syphilitic involvement of the aortic valve, was the result of adhesions between the cusps of the aorta and the corresponding aortic wall. The widening of the commissures, therefore, cannot be explained on a mechanical, but must be explained on an inflammatory basis.

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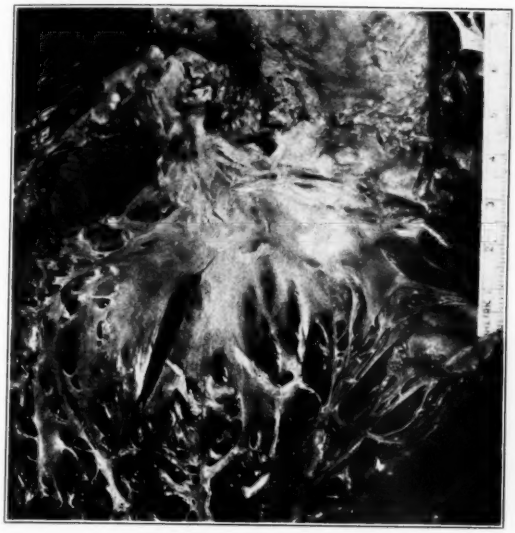
DESCRIPTION OF PLATE

PLATE 66

FIG. 1. Heart of Case 1. Note the slit-like opening of the sinus of Valsalva, corresponding to the posterior cusp.

FIG. 2. Portion of the aorta and aortic valve of Case 1.



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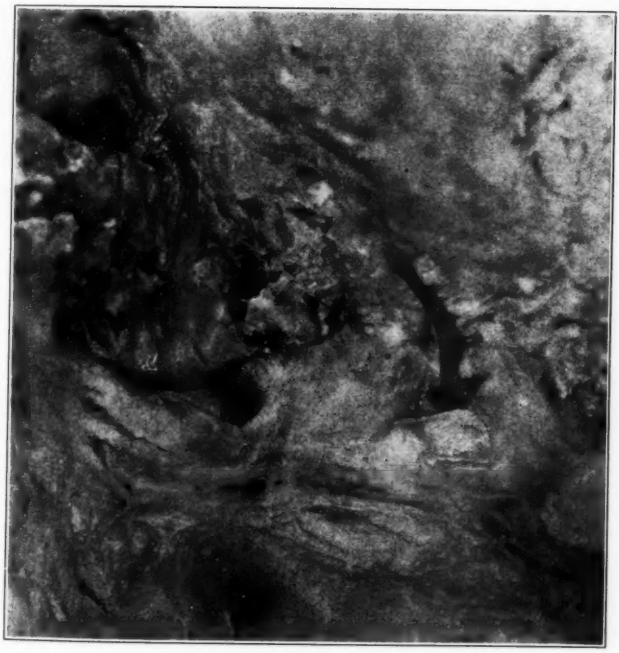


2

Saphir and Stasney

Aortic Valve in Syphilitic Aortitis

PLATE 67

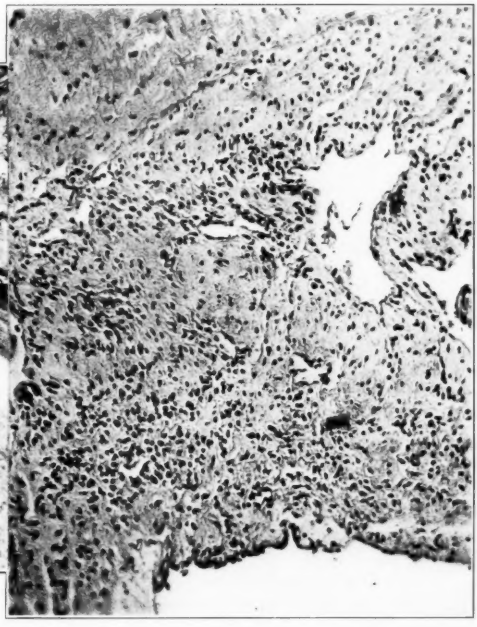
FIG. 3. Region of the commissure of the aortic valve of Case 1. Note the newly formed blood vessels and the lymphocytic infiltration. Iron hematoxylin-orcein stain. × 110.

FIG. 4. Base of the aortic valve of Case 1. Note the newly formed blood vessels and lymphocytic cells. Iron hematoxylin-orcein stain. × 200.

FIG. 5. Slit-like opening of the sinus of Valsalva in Case 1. Hematoxylin-eosin stain. × 110.

FIG. 6. Aorta and aortic valve of Case 2 cut longitudinally. Note the collection of blood in the sinus space.

3



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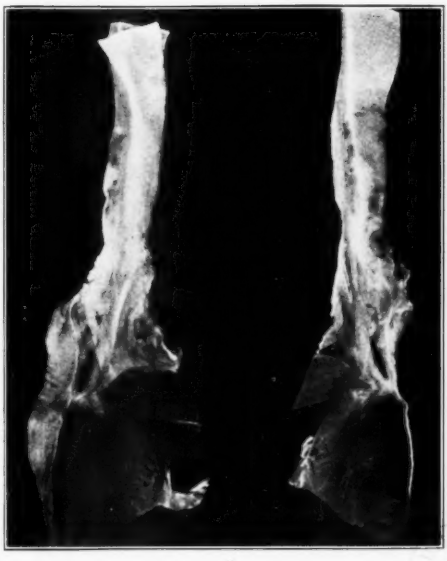

6

Saphir and Stasney

Aortic Valve in Syphilitic Aortitis

RHEUMATIC HEART DISEASE WITHOUT VALVULITIS*

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The case of rheumatic heart disease to be reported is of unusual interest because of the absence of specific lesions of the heart valves and because an anatomical diagnosis of rheumatic fever depends on the presence of lesions that have only within late years been considered as characteristic of this disease.

In his review Sacks [1] says: "There have been but few reports of cases of proved rheumatic myocarditis in which the valves were entirely free of either recent or old disease. Aschoff's case [2] of diffuse interstitial myocarditis and Denzer's case [3] are examples." Sacks also mentions three cases (Geipel,[4] Fraenkel,[5] Libman [6]) in which endocarditis was absent in hearts exhibiting pericarditis accompanied by Aschoff bodies in the myocardium. We have been able to find reports of additional cases in which the valves were said to have been normal, and undoubtedly other cases exist. Following is a brief résumé of these cases.

Aschoff's Case [2] *(1904):* Death came suddenly three weeks after a hand wound, which meanwhile had healed. Autopsy demonstrated an acute interstitial myocarditis with no sign of endocarditis. Organisms did not grow in cultures and were not demonstrable in sections of heart muscle. The heart muscle showed some necrosis and was infiltrated by many eosinophils and by some adventitial cells, plasma cells and lymphocytes. The most noteworthy thing, however, was the presence of a few characteristic nodules composed of large cells and having an appearance similar to nodules described in cases of rheumatic myocarditis. Such nodules, submiliary in size, lay in close relation to small or medium sized vessels and were composed of extraordinarily large, closely approximated cells having one or more large, slightly indented or irregular shaped nuclei. The centers of the nodules were often formed of poorly staining or apparently necrotic masses of fused cytoplasm.

Geipel's Case [4] *(1905):* Male, aged 17 years. There was a history of pain in the joints and dyspnea. Autopsy showed fibrinous pericarditis, bilateral fibrinous pleural exudate, edema of the lungs and chronic passive congestion of the viscera. The heart valves were entirely delicate and showed no vegetations. Aschoff bodies were present in the myocardium.

Geipel's Case [7] *(1907, 1909):* Stone cutter, aged 53 years. There was a history of pain in the joints. After a long walk the patient suddenly fell dead on the street. Autopsy showed great hypertrophy and dilatation of the right ven-

* Received for publication March 27, 1933.

tricle. Microscopically the myocardium showed Aschoff bodies and a diffuse interstitial myocarditis. The coronary arteries were characteristically involved.

Fraenkel's Case [5] (*1912*): Female, aged 21 years. History not available. Autopsy showed obliterative pericarditis. Aschoff bodies were found in the myocardium. Streptococci were present in blood culture.

Poynton [8] (*1918*): In discussing rheumatic pericarditis Poynton stated that endocarditis is almost an invariable accompaniment, "though occasionally we find no evidence of its occurrence."

Libman's Case [6] (*1923*): Libman reports: "We have observed one case in which there was a recent pericarditis accompanied by Aschoff bodies in the heart muscle, but not by any lesions of the valves."

Denzer's Case [3] (*1924*): Female, aged 23 months. Death was due to cardiac failure 5 days after the onset of symptoms. White blood corpuscles 20,000. Temperature did not rise above 100° F. Blood culture sterile. Autopsy (performed by Sacks) showed an enlarged heart with a few hemorrhagic areas on the visceral pericardium. The tricuspid, pulmonary and aortic valves were normal. The mitral valve was not narrowed, its edges were somewhat thickened but there was no evidence of either past or present endocarditis. The left ventricle was hypertrophied and the muscle was flabby and showed tigering. Microscopic examination of the heart muscle showed Aschoff bodies. The heart muscle was edematous and occasional areas of degeneration with cellular infiltration were found.

Swift's Cases [9] (*1924*): Swift reports: "Recently, two fatal cases have been brought to our attention in which myocardial weakness was the sole clinical picture, and postmortem the only distinct lesions were Aschoff bodies widely disseminated throughout the heart muscle."

VonGlahn and Pappenheimer: VonGlahn [10] (1927) in a study of 109 cases of rheumatic disease of the heart, noted that the myocardium was involved in 8 cases where the valves and pericardium were normal. VonGlahn and Pappenheimer [11] (1926) have published data of 2 of these cases.

CASE 1: Male, aged 68 years. There was no history of tonsillitis, arthritis or chorea. Wasserman reaction negative. Past history suggested myocardial insufficiency. Autopsy showed acute rheumatic myocarditis with Aschoff bodies present, thrombi in the right auricle and the right and left ventricles, emboli in the right internal carotid and right middle cerebral arteries, infarct of the brain and rheumatic lesions in the blood vessels of the testes and pancreas.

CASE 2: Female, aged 33 years. There was a history of tonsillitis but no history of arthritis, chorea or cardiac trouble. Autopsy revealed rheumatic myocarditis with Aschoff bodies present, rheumatic lesions in blood vessels of the ovary and kidney, chronic nephritis and hypertrophy of the heart.

Chiari's Case [12] (*1928*): Female, aged 29 years. There was a history of "heart trouble" over a period of several years and of shifting pain in the joints beginning 6 months before death. Death was due to heart failure. Autopsy showed a fresh fibrinopurulent pericarditis, cloudy swelling of myocardium, liver, and kidneys, and acute splenic tumor. Microscopic examination revealed Aschoff bodies in the myocardium and characteristic rheumatic lesions in the aorta.

Of considerable importance are the findings of Swift,[13] Holsti,[14] and Kugel and Epstein [15] that valvulitis may be present in leaflets that grossly appear normal or little altered. One can only speculate

as to the possibility of microscopic changes having been present in the valves in some of the cases reviewed above. Valvulitis is probably early accompanied by slight gross thickening of valve leaflets, and it is not likely that such thickening would be overlooked by one making a diagnosis of rheumatic myocarditis when verrucae were absent from all the valves.

Report of Case

Clinical History: H–22483. A Chinese girl, S. H. S., aged 12 years, was brought to the Peiping Union Medical College Hospital on Nov. 30, 1928, complaining of shortness of breath, palpitation of the heart, cough, and swelling of the lower extremities and abdomen. In the past she had frequently had sore throat, but there was no history of pain in the joints, long standing fever or severe illness.

The onset of the present illness began with edema of the ankles in June, 1927. A month later the abdomen became swollen and the patient experienced palpitation of the heart and dyspnea. Some fever and "pain in the bones" of the extremities were said to have been present at that time. Two long hospitalizations brought only temporary improvement.

Physical examination showed a well developed and well nourished girl, dyspneic and slightly cyanotic. Venous pulsation was visible in the neck. The heart was markedly enlarged and there was a well marked precordial heave. A systolic thrill could be felt at the apex. The second sound was louder at the pulmonic than at the aortic area. Pulse rate 90 to 96. Blood pressure 108 systolic and 72 diastolic. The liver was markedly enlarged. There was moderate pitting edema of the legs and ankles, and a suggestive fluid wave in the abdomen was obtained.

The urine contained a faint trace of albumin and a few granular casts. Blood examination showed a mild degree of anemia. Electrocardiogram taken at the time of admission showed normal mechanism.

Seven weeks hospitalization brought moderate improvement in the general condition. Symptoms reappeared, however, under light work at home and the patient returned to the outpatient department. Digitalis was prescribed. By mistake she took an overdose, and 4 days later, on Feb. 8, 1930, she was readmitted to the hospital with auricular fibrillation and signs and symptoms more marked than noted at the previous admission. A few squeaking râles were heard at the back of the chest. The temperature was 39.9° C, but later remained normal or subnormal. The pulse rate was 45 and the rhythm was irregular. Blood pressure 105 systolic and 50 diastolic. The white blood cells numbered 12,650, of which the proportion of polymorphonuclear neutrophils was 49 per cent, lymphocytes 44 per cent, large mononuclears 6 per cent, and eosinophils 1 per cent. In the hospital the patient complained of severe pain in the legs. Salyrgan was given several times with marked transitory diuresis following each injection. On account of auricular fibrillation quinidine was given on February 19. The drug was given in three doses at 1½ hour intervals, the first two doses being 0.1 gm. each, and the third dose 0.3 gm. Twenty minutes after the third dose the patient collapsed and died 6 hours later.

The clinical diagnoses were: rheumatic cardiovascular disease, mitral stenosis and insufficiency, cardiac enlargement, cardiac insufficiency, partial heart block, auricular fibrillation, bronchitis and cardiac failure following quinidine therapy.

Postmortem Examination

Autopsy was performed 17 hours after death. The body was that of a fairly well developed and fairly well nourished Chinese girl weighing 28 Kg. and measuring 137 cm. in length. The vessels of the conjunctival mucous membranes were overdistended with blood. Thick mucus adhered to the nasal septum. The cervical lymph nodes were readily palpable. The nail beds were blue. The thorax flared slightly at the lower border and the abdomen was rounded and much distended. There was no obvious edema of the lower extremities.

The abdominal cavity contained between $2\frac{1}{2}$ to 3 liters of almost clear, amber-colored, watery fluid. The lower border of the liver extended 3 cm. below the costal margin in the right midaxillary line. The intestines contained but little material.

The heart occupied a relatively large volume of the thoracic cavity. The pericardium surrounded the heart rather tightly and was not thickened. There were no pericardial or pleural adhesions and the surfaces were smooth and glistening.

The heart was greatly enlarged, weighing 360 gm. The right auricle and ventricle were much dilated. Anteriorly, near the apex of the right ventricle, the epicardium exhibited a slightly elevated, grayish white area of fibrous tissue thickening. Projecting from the epicardium of the left auricle was a soft, rounded, thick mass 4 mm. in diameter of smoothly outlined, fibrous tissue growth. A few petechial hemorrhages were noted in the epicardium of the left ventricle. The heart chambers contained a soft purple clot, also a jelly-like, chicken fat clot. Thrombi were looked for carefully in all the heart chambers and were not found. The foramen ovale and the ductus arteriosus were closed. The right auricle was dilated so that the trabeculae carneae were conspicuous; the wall in the areas between the trabeculae was quite thin and was semitransparent. The tricuspid orifice admitted three fingers, and the valve circumference measured 11 cm. The valve leaflets were a little diffusely thickened, but were quite pliable and free from vegetations. The dilatation of the right ventricle was marked, and the ventricle wall

was hypertrophied to a thickness of 4 to 6 mm., being thickest just below the pulmonic valve. The pulmonic leaflets were delicate and the valve orifice measured 7 cm. in circumference.

The left auricle was much hypertrophied and dilated. The auricular appendage was, however, normal in size. The endocardium of the right lateral wall of the auricle showed a corrugated, slightly elevated (about 1 mm.), poorly outlined patch 5 or 6 cm. in area. Except for the presence of a number of small, pinkish red mottlings, the color of the patch did not differ from that of the normal endocardium. Opposite this area the previously noted mass of soft fibrous tissue took its origin from the epicardium. Anteriorly and inferiorly to the patch described, the endocardium was slightly thickened and showed several smooth shallow depressions up to 2 mm. in diameter and 1 mm. deep. Over one small area the auricle wall was thin and fibrous. The mitral orifice was abnormally wide, admitting three fingers, and had a circumference of 12 cm. The valve leaflets and some of the chordae tendineae were a little thickened, but the leaflets were quite flexible and were not shrunken. There were no vegetations or nodular thickenings on the leaflets.

The left ventricle was hypertrophied and moderately dilated so that the apex was rather rounded. The wall varied in thickness from 16 mm. at the base to 11 mm. at the apex. The endocardium was slightly thickened and had a gray color below the aortic valve and over the trabeculae near the apex. The aortic orifice measured 6.5 cm. in circumference. The leaflets were delicate. The myocardium was flabby, and the cut surface had a glistening, light brownish pink color with some delicate mottlings of yellow and pink and a few small, opaque, gray, linear markings. The large coronary arteries were normal in appearance.

Near the sinuses of Valsalva and in the abdominal portion the aorta presented small, delicate, barely visible grayish yellow mottlings. The pulmonary artery and its large branches showed elevated grayish yellow plaques. Iliac, common carotid, splenic and mesenteric arteries and the celiac axis appeared to be normal.

The lungs, particularly the right, were a little heavier and firmer than normal. The large bronchi contained thick mucus and foamy mucoid fluid. Tracheobronchial lymph nodes were enlarged, and their cut surfaces were a velvety reddish black. One lymph node showed grayish areas judged to be tubercles. The lung cut surfaces

were brownish red and pink. Scattered in both lungs, but found more abundantly in the right and especially in the middle lobe, there were small, slightly elevated, dark red, solid areas. Pressure caused the escape of a little foamy fluid from the cut surface of the left lung, and a considerable amount in the case of the right lung.

The liver was much enlarged and weighed 1200 gm. The cut surface showed nutmeg-like, reddish purple markings and in places the lobule architecture could not be made out. The spleen was enlarged to weigh 235 gm., was firm, and presented a pinkish red, slightly velvety cut surface with inconspicuous trabeculae and readily visible malpighian bodies. The intestinal veins contained blood, and the intestinal mucosa was a grayish red color. The stomach mucosa was a dark red color and was covered by thick mucus. Mesenteric and pancreatico-duodenal lymph nodes were somewhat enlarged. The kidneys were rather firm and weighed 160 and 140 gm. respectively. The capsules stripped easily, revealing smooth, purplish red surfaces. Cut surfaces were darker red than normal, striations were straight, and malpighian bodies were conspicuous. The cortex of the suprarenal glands was not as yellow in color as normal. The tonsils were small and not remarkable in gross. Paratracheal and deep cervical lymph nodes were a little enlarged, but cut surfaces were not unusual. Most of the supporting soft tissues of the body were abnormally moist.

The brain appeared anemic. In the right carotid artery, at the point of its division to form vessels of the brain, there was an obstructing mass that was not readily removed. Mucosa of sphenoid and maxillary sinuses was a little thickened and abnormally moist. Turbinate mucosa, ethmoid and mastoid cells, and middle ears appeared normal.

Microscopic Examination

Tissues were fixed in Zenker-formol and in 10 per cent formalin, and later several blocks were cut from tissues fixed in Kaiserling fluid. Sections were stained with hematoxylin and eosin, except as otherwise noted.

Heart: In four sections through the left ventricle, the muscle striations stain rather poorly, and many of the cells appear shrunken and degenerated. Some fragmentation is present. A scharlach R

stained frozen section reveals only a small amount of fat in the muscle cells, and this is diffusely distributed. In some areas the connective tissue of the epicardium and endocardium and that between shrunken muscle cells contains a few small round cells resembling lymphocytes, some large mononuclear wandering cells, and a few polymorphonuclear leukocytes. In a section taken through the patch of thickened epicardium, noted in gross on the right ventricle, the structure is seen to be composed of loosely woven, highly vascularized connective tissue sparsely infiltrated by small round cells. Another section taken through the right ventricle shows several small areas of muscle cell degeneration with fibrous tissue proliferation adjacent. In the two sections taken the posterior leaflet of the mitral valve shows no lesions.

Five blocks were taken through the left auricle in the region of, or adjacent to, the corrugated patch. The endocardium in all these sections is distorted and thickened by connective tissue growth, and together with the adjacent muscle layer shows wandering cell infiltration and usually edema. In the sections through the corrugated patch, thickening of endocardium by loosely woven connective tissue is marked, and varies in degree to make the surface quite irregular in outline. In certain areas fibroblasts are numerous; in other more edematous areas the young connective tissue cells are undergoing degeneration with vacuolization of cytoplasm and karyolysis. A lining of endothelial cells is recognized over most of the endocardium, but one section shows several areas of old and fresh blood and fibrin in crypts apparently formed by closely approximated folds of thickened endocardium. Numerous large mononuclear wandering cells, numerous small round cells resembling lymphocytes, and a few polymorphonuclear leukocytes diffusely infiltrate parts of the newly formed endocardium and are most abundant in the lower part of the edematous endocardium adjacent to the auricle muscle. Blood capillaries are conspicuous in this lower layer of endocardium and in near-by muscle, and the cellular infiltration is often marked about these vessels. There is also a tendency for the infiltrating cells to be arranged in long, thick and thin rows parallel with the surface, these being apparently lines of separation in the connective tissue. The interstitial tissue of the muscle layer is edematous, and many of the muscle cells are poorly preserved. Hypertrophy of muscle cells is marked. In some areas, especially about blood vessels, muscle cells

have been replaced by loosely woven fibrous tissue, and this tissue holds a few small round cells and mononuclear wandering cells. In several of the sections the epicardium is thickened by edematous fibrous tissue containing dilated capillaries, a moderate number of small round cells and large mononuclear wandering cells. The soft epicardial growth opposite the corrugated patch is seen to be a highly vascularized, edematous growth of fibrous tissue infiltrated by many inflammatory cells of the types just mentioned.

Aschoff bodies were looked for carefully in all the sections but were not found. Some of the large mononuclear cells in the left auricle exudate resemble Aschoff cells in appearance, and with the methyl green-pyronin stain the cytoplasm of a few such cells stains red. The stain could not be said to be satisfactory, however, with Zenker-formol-fixed material. The coronary arteries appear normal. Van Gieson and Mallory connective tissue stains demonstrate well the increase in connective tissue in the auricle endocardium, epicardium and interstitial muscle tissue. Verhoeff elastic tissue stain shows elastic fibers in some of the newly formed connective tissue of the auricle endocardium. With the MacCallum bacteria stain and with the Levaditi stain no organisms are demonstrated.

Aorta: In some of the four sections the intima is a little thickened by loosely woven or moderately dense fibrous tissue. In places the connective tissue cell nuclei are more thickly distributed than in normal intima, and a large mononuclear wandering cell is occasionally seen. Fat stain shows minute fat globules in the cytoplasm of some of the intimal connective tissue cells. Parts of the media appear to be edematous, and there are large linear areas where normal tissue is replaced by delicate connective tissue fibers. Loss of tissue is also evident about some of the vasa vasorum. The Verhoeff-Van Gieson combination stain shows the loss of elastic tissue in parts of the media, and this change is greater in the half of the media nearest the intima. The internal elastic lamella in places shows poorly. In the adventitia of two of the sections, and of one in particular, there is moderate perivascular and diffuse infiltration by large mononuclear wandering cells, a few plasma cells and a few small round cells resembling lymphocytes.

Pulmonary Artery: Sections taken through the grayish yellow plaques show great thickening of intima by loosely woven fibrous tissue, and many cells contain fat. The media appears to be slightly

edematous, the elements being spread apart to a greater extent than normal. The Verhoeff-Van Gieson stain demonstrates diminution of elastic tissue fibers, not only adjacent to the intima but also to a slight extent in other parts of the media. Recognizable, partially disintegrated elastic tissue fibers are seen in some of the linear areas not taking the black stain. The adventitia in one of the sections shows large mononuclear wandering cells and small round cells, perivascularly and diffusely distributed.

Other Vessels: In the pulmonary vein and in the several medium-sized arteries and veins examined, the only thing found worthy of note is slight intimal thickening in the superior mesenteric artery. In the section through the right internal carotid artery just below its point of bifurcation the lumen is filled by a laminated structure composed of poorly preserved fibrin, red blood cells, nuclear débris and material that appears to be disintegrated blood platelets. At the edge of the mass there is well preserved fibrin enmeshing intact blood cells. The mass is not attached to the intima of the artery.

Other Organs: The lung alveolar wall capillaries are dilated, and a little thickened by fibrous tissue. Many alveoli contain coagulated fluid, red blood cells and large mononuclear cells, some of the latter containing much brown pigment. In areas alveoli contain coagulated fluid, fibrin and polymorphonuclear leukocytes. In a section from the left lung a few collapsed alveoli are present. The mucosa of a main bronchus contains a moderate number of lymphocytes and a few polymorphonuclear eosinophils. In a bronchial lymph node blood capillaries are conspicuous and a few epithelioid cell tubercles are to be seen; in another section the sinuses contain a few red blood cells and large mononuclear wandering cells with a polymorphonuclear leukocyte occasionally visible.

The sinuses of the spleen and the liver are greatly distended with blood. The splenic connective tissue is slightly and generally increased. Liver cells about many of the efferent veins are atrophied, and contain fat and bile pigment. Half a dozen well formed miliary tubercles are noted.

In the kidneys the capillaries are tremendously dilated. Many capsular spaces contain finely granular material and there appear to be fibrin adhesions between a few glomeruli and their capsules. The tubule epithelium stains poorly and exhibits swelling, partial disintegration and karyolysis. Many tubules contain hya-

line and mononuclear cell casts, and finely granular material. A small amount of fat can be demonstrated in the tubule cells. Interstitial connective tissue is increased diffusely to a slight extent, and to a considerable extent in some areas where leukocytes are present. In sections of stomach and intestine blood capillaries are greatly dilated. No parasites or ova are found in the intestinal contents. Cortical cells of the suprarenal glands contain but little lipoid. No *Treponema pallida* are found in a Levaditi-stained section of suprarenal gland.

The tonsils show small fibrous tissue scars and numerous dilated crypts containing epithelial cell débris and masses of bacteria. Polymorphonuclear leukocytes are found in a few crypts and also in their epithelial cell walls. The MacCallum bacteria stain reveals in the crypts numerous Gram-positive cocci, often in chain formation. In the large masses of organisms, bacilli of several types outnumber the cocci. In two sections from nasal sinuses the mucosa is edematous and infiltrated to a greater extent than normal with lymphocytes and larger mononuclear wandering cells. Some polymorphonuclear leukocytes are present. Gram-stained smears from the sphenoid sinus show a few polymorphonuclear leukocytes and numerous Gram-positive cocci in pairs and chains. Sections of thyroid, thymus, parathyroid, hypophysis, ovary, uvula, esophagus, turbinates, urinary bladder, pancreas, voluntary muscle, femur bone marrow, and brain present nothing particularly noteworthy.

Bacteriological Examination: Culture of heart's blood in broth, and cultures of heart's blood and of spleen tissue on blood agar plates showed no growth after 48 hours incubation.

Anatomical Diagnoses: Rheumatic fever with lesions in endocardium (left auricle), myocardium, epicardium, aorta and pulmonary artery; cardiac hypertrophy and dilatation; fibrous epicardial plaques; mitral insufficiency; arteriosclerosis of pulmonary artery; marked chronic passive congestion of viscera; ascites; anasarca; edema of lungs (slight); partial atelectasis (left). Chronic bronchitis; early lobular pneumonia; chronic lymphadenitis of bronchial, tracheal and cervical lymph nodes. Hyperplasia of mesenteric lymph nodes. Embolus in right internal carotid artery. Slight subacute nephritis. Chronic tonsillitis. Slight sphenoid and maxillary sinusitis. Tuberculous lymphadenitis of bronchial lymph node; miliary tubercles in liver. Accessory spleen.

Discussion

The absence of Aschoff bodies in this case does not preclude the diagnosis of rheumatic fever. In 190 cases of rheumatic fever reviewed by Clawson [16] Aschoff bodies were found in only 67 per cent of the cases. Thayer's [17] included series of 24 cases showed the highest reported incidence of Aschoff bodies, 87.5 per cent. In 24 picked, active cases Kugel and Epstein [15] reported Aschoff bodies in 79.2 per cent; and in 28 picked active cases McClenahan and Paul [18] reported Aschoff bodies in 85 per cent. The last two sets of figures offer additional support to Clawson's statement that "the more acute the disease process the greater is the likelihood of Aschoff bodies being present." The case reported here was chronic in character.

The lesion in the left auricle in this case was the most important single piece of evidence leading to the diagnosis of rheumatic fever. MacCallum [19] first described the auricle lesions characteristic of this disease. VonGlahn [20] confirmed the finding, and reported characteristic auricle involvement in 9 of 31 cases examined. In Thayer's series [17] of 25 cases the left auricle was involved in 10 cases. Kugel and Epstein [15] record auricle lesions in 16 of 24 cases. When these figures are combined there is record of auricle involvement in 35 of 80 cases (44 per cent). Clawson [16] reports: "Microscopic evidence of infection in the auricle was found almost constantly."

Characteristic changes in the aorta in rheumatic fever were first recognized by Klotz.[21] Pappenheimer and VonGlahn [22, 23, 24] described and illustrated several stages of the lesions in all three layers of the aorta. Perla and Deutch [25] added the description of an additional acute lesion consisting of fibrin plaque formation in the intima. In 77 cases examined in which there was presumptive evidence of recent or old rheumatic infection, Pappenheimer [26] reported triangular or flame-shaped scars in addition to acute lesions in 40 per cent of the cases. Kugel and Epstein [15] recorded 5 cases with aortic involvement (diffuse inflammatory reaction in the media) in a series of 24 cases of active rheumatic fever. Chiari [12] reported characteristic adventitial changes (with normal media) in 5 of 6 cases where death was due to an acute exacerbation of rheumatic fever, and expressed doubt as to whether the 6th case was really rheumatic fever, inasmuch as Aschoff bodies were not found. Giraldi [27]

found characteristic microscopic lesions of the aorta in all of 5 consecutive cases of acute rheumatic fever. Klinge and Vaubel [28] described the aortic changes in a series of cases of rheumatic fever and recognized three stages in the life history of a lesion.

The changes of the aorta in the case here reported do not fit the picture of syphilis or of medionecrosis aortae, as described by Gsell, by Erdheim, and by Cellina. Aortitis occurs in various diseases in which bacteria circulate in the blood — focal reaction, necrosis and thrombosis are features of such — but, as Siegmund [29] has shown, the picture is not necessarily completely developed. Necrosis of media has been described in infectious diseases, notably in scarlatina, diphtheria and typhoid fever. Because the aortic lesion in this case was in a rather inactive stage and showed no specific cellular reaction, one should be open-minded with regard to its interpretation. However, considering all the facts in the case, the lesion is best explained as being related to rheumatic fever. As in the first cases described by Pappenheimer and VonGlahn [22] and a case more recently described by Gray and Aitken [30] (Case 3), this case showed scarring of the media as the most striking alteration. The cellular infiltration and the fibrous tissue thickening of the intima were slight but perfectly definite.

Lesions similar to those found in the aorta in rheumatic fever have been described in the pulmonary artery by Paul,[31] Kugel and Epstein [15] (5 instances in 24 cases), Gray and Aitken,[30] Shaw,[32] and Chiari.[33] In the case here described there was moderate cellular infiltration in the adventitia of the pulmonary artery, the media appeared to be edematous and showed degeneration of elastic tissue fibers. Because of the marked arteriosclerotic change in the intima one probably is not justified in emphasizing the rheumatic character of the rather slight lesions in the media, but they fit in with the diagnosis of rheumatic fever as made on the basis of other evidence in the case.

The heart was hypertrophied and greatly dilated to the extent that beriberi was at first considered as a possibility. The mitral ring was enlarged to produce a relative mitral insufficiency. Long-standing passive congestion of the pulmonary circulation was evidenced by the arteriosclerosis of the pulmonary artery and by the fibrosis and pigmentation of the lungs. Marked chronic passive congestion of abdominal viscera, ascites and anasarca were signs of myocardial

insufficiency. The degree of myocarditis of the ventricles was not great, but there were moderate diffuse fibrosis and degenerative change in the muscle. In the left auricle the endocarditis and myocarditis were marked. Was there at one time a slight valvulitis of the mitral leaflets? If so, no gross or microscopic evidence remains to prove it. Scars in the auricle extend rather close to the valve ring. The slight thickening of the mitral leaflets and of some of the chordae tendineae could be related to the increased strain to which they must have been subjected because of the imperfect closure of the valve. The autopsy findings correlate well with the history that the first signs of congestive heart failure appeared 1 year and 8 months before death.

The embolus in the right internal carotid artery explains the mode of death. Such an occlusion would not be fatal in an individual with good blood circulation, but it would be expected to be so in one having a decompensated heart. Although no thrombi were found in the heart, the presence of disintegrating blood and fibrin in the endocardial lesion of the left auricle makes it the likely site of thrombus formation. It is interesting to note that Willius,[34] in an analysis of the mode of death in 160 cases of rheumatic heart disease, reported death from cerebral embolism in 4 cases, and one other death from coronary occlusion by an embolus derived from a thrombus in the left ventricle. There were thrombi in the auricular appendages in 5 of Thayer's[17] 25 cases. In 1 of the above noted cases of Von Glahn and Pappenheimer[11] without valve lesions death was due to cerebral embolism. It is evident that cerebral embolism may be expected occasionally as a terminal event in rheumatic heart disease. In our case the short time between the administration of quinidine and death leads to the belief that quinidine effect was responsible for dislodging a thrombus from the auricle. Eismayer[35] collected records of 12 instances of embolism in 934 cases of heart disease treated by quinidine; 16 other deaths in his series were apparently due to the direct effect of the drug.

Summary

In the literature there have been noted 17 cases in which the diagnosis of rheumatic fever was made at autopsy and in which all the heart valves were apparently free from rheumatic lesions; all of these

cases exhibited myocarditis with Aschoff bodies present, and in 4 cases pericarditis also was present.

Based on criteria as we know them at the present time, there is reported an additional case of chronic rheumatic fever in which the heart valves were free from rheumatic lesions. Aschoff bodies were not found and the diagnosis of rheumatic fever depended on the presence of characteristic lesions in the left auricle, scars in the media of the aorta, slight changes in the media of the pulmonary artery, and diffuse inflammatory cell infiltrations in the heart and in the adventitia of the aorta and the pulmonary artery. Death came 1 year and 8 months after the first symptoms of congestive heart failure, following the administration of quinidine. An embolus, accounting for the mode of death, was found in the right internal carotid artery and is believed to have been formed on the rheumatic endocardial lesion in the left auricle.

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DESCRIPTION OF PLATES

PLATE 68

FIG. 1. Heart. Hypertrophy and dilatation are apparent. Corrugated areas of thickened endocardium are visible near the lateral edges of the auricle, on the right side of which are a number of crater-like scars of healed endocardial lesions. The auricle wall near-by is thin. The mitral ring is enlarged to produce relative insufficiency of the valve. Except for slight thickening the valve leaflets are normal. Small, polypoid fibrous tissue growths projecting from the auricle are visible in the upper left-hand corner of the photograph.

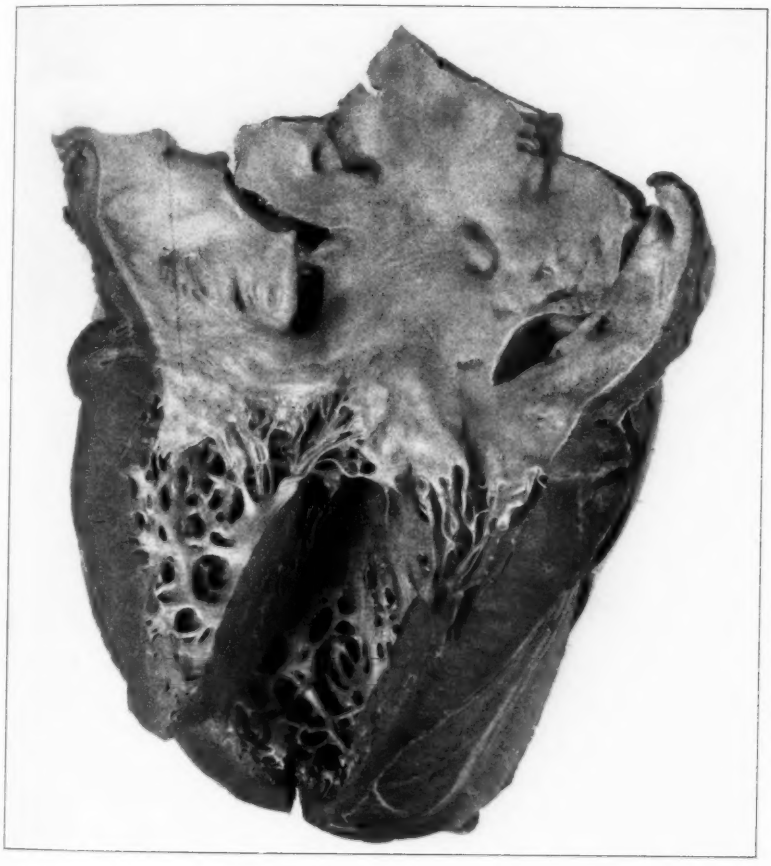

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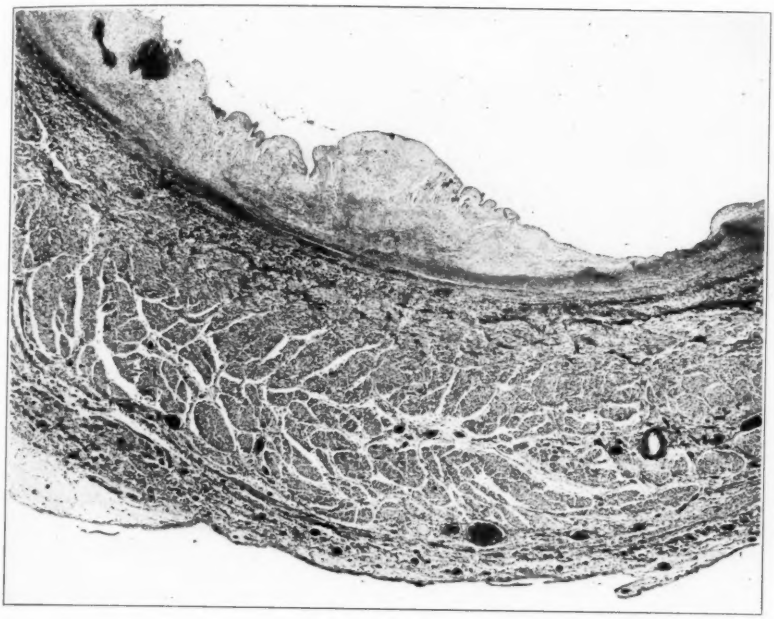
PLATE 69

FIG. 2. Left Auricle. Hematoxylin and eosin stain. The section was taken through the corrugated patch and shows irregular fibrous tissue thickening of the endocardium, a great number of infiltrating inflammatory cells (visible merely as black dots), some fibrosis of the muscle layer and slight thickening of the epicardium. The two dark patches in the endocardium at the upper left represent areas of thrombus formation. × 15.

FIG. 3. Aorta. Verhoeff-Van Gieson stain. The elastic tissue fibers of the media are much diminished and the media architecture is distorted. The intima is a little thickened by fibrous tissue. × 90.



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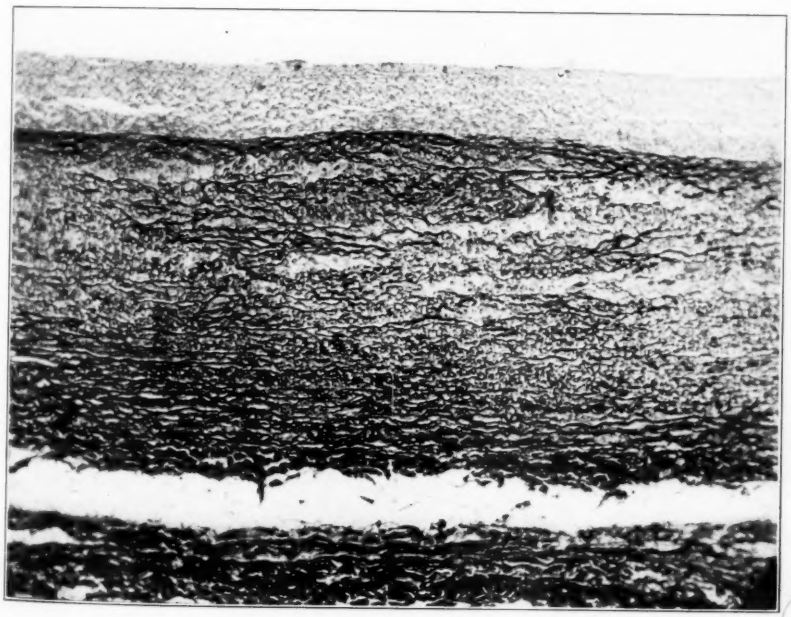

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Garber

Rheumatic Heart Disease without Valvulitis

THE PERCENTAGE OF THE DIFFERENT TYPES OF CELLS IN THE ANTERIOR LOBE OF THE HYPOPHYSIS IN THE ADULT HUMAN FEMALE *

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INTRODUCTION

The necessity of greater accuracy in the evaluation of the relative number of the different types of cells regularly recognized in the anterior lobe of the hypophysis was briefly discussed in a previous report [1] that dealt with a differential count of these cells in 111 normal male adults. The emphasis recently placed on basophilic adenomas of the pituitary (Cushing,[2] Moehlig [3] and others), and the suggestion that profound abnormalities (painful adiposity, kyphosis, sexual dystrophy, and so on) may result from comparatively small masses of basophilic cells, serve further to emphasize the point.

Acidophil and *basophil* are used instead of the more exact terms *alpha* and *beta*, proposed by Bailey and Davidoff,[4] because of more common usage. Other designations for these chromophilic cells, as well as for the chromophobes, will be found in a preceding article.[1] The section on the hypophysis by Bucy,[5] recently published, may further be cited for the normal cytology.

On account of the close interrelations of this organ with the gonads, a sex difference in the histological structure is of some interest. It has been recognized for some time that in some animals (rat, rabbit, cattle), as well as in the human, the female has a larger hypophysis, due to a larger anterior lobe. Further corroboration of this for the human on the basis of quantitative determinations on over 200 selected cases will be published in the near future. There are, however, no very reliable data showing which of the elements in the anterior lobe is responsible for this difference in size. A recent examination of serial sections of 235 normal, adult human hypophy-

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ses [6] indicates that gland-like extensions of pars intermedia into the neural lobe is much more frequent in the female than in the male. The incidence of concretions resembling "brain-sand" in the pial investment of the upper aspect of the normal hypophysis was found to be nearly twice as high in females as in males. Neither pregnancy nor age seems to have any relation to the frequency of these two structural details.

Much has been written on the development of a special cell in the anterior lobe of the hypophysis during pregnancy in the human and in certain other animals. Descriptions of these so-called "pregnancy cells" are so contradictory and vary so much (Severinghaus, Engle and Smith [7] and Stein [8]) that considerable attention has been paid to the identification of such cells in cases of sudden and accidental death of pregnant females. Interest along this line has been heightened by the apparent overproduction of gonad-stimulating hormones by the anterior lobe of the hypophysis during pregnancy (the basis of the Aschheim-Zondek [9] test for pregnancy).[10] While one might anticipate under these circumstances that there would be recognizable histological changes having a bearing on the specific function of the different types of cells, it must also be remembered that cells that are apparently similar morphologically, if not actually identical, may produce several hormones, just as the cells of the pancreatic alveoli, for example, produce several ferments.

These considerations are the justification for the establishment of norms toward which it is hoped these data will contribute.

Method

The histological technique has been uniform throughout and is the same as that used on the male hypophysis.[1] It consists essentially of formalin fixation and staining, first slightly with hematoxylin (to bring out the nucleus, which is important in counting cells so variable in size), and then (after washing in tap or alkaline water followed by distilled water) in Mallory's connective tissue stain (acid fuchsin followed by a mixture of orange G and anilin blue) as in the usual procedure for collagen fibers (Mallory[11]), with the precaution that the orange G-anilin blue stain must be washed off in not lower than 95 per cent alcohol and dehydrated rapidly in absolute alcohol. To bring out most strikingly the basophilic cells, omit the acid fuchsin

and stain only in the orange G-anilin blue mixture. By leaving out the acid fuchsin differentiation between the acidophilic cells and the chromophobes is not so marked. This is a far simpler technique than that proposed by Bailey [12] and adequate for general purposes where mitochondria and other finer cytological features are not required. The colored frontispiece of Cooper's book [13] is no exaggeration of the brilliancy obtainable, even on material obtained twelve or more hours after death. It works fully as well after saturated corrosive sublimate containing 10 per cent formalin, and Howes [14] finds it the most effective stain, particularly after Gilson's fixation (a corrosive sublimate mixture). If neutral formalin is used as the original fixing agent and either the block or the sections mordanted in potassium dichromate, finer cytological details may be brought out by mitochondrial and other stains. As Bailey [15] again points out, the hematoxylin and eosin stain is not differential enough.

If such a small tumor as was present in the Bauer case (cited by Cushing[2]) is of as much consequence as that and some other cases suggest, it is to be hoped that in all suspicious cases of hypophyseal involvement, whether there are any gross indications in the hypophysis or not, the complete gland will be sectioned serially and thirty or more sections at equal intervals stained. If cut 10 microns thick, every twentieth section is enough. These can usually be mounted on four ordinary slides and hence involve very little extra work. For quantitative evaluation of the number of cells thinner sections are necessary.

Kernohan [16] has found that ordinary formalin-fixed human hypophysis may be treated easily so as to stain differentially also by Heidenhain's modification of Mallory's stain. The basophils are very prominent by this method. Biedermann [17] used still another combination of stains that differentiates well the three types of cells after formalin-fixation, and Soós and Csizek [18] have recently reported that a resorcin-fuchsin stain is highly selective for the basophilic granules, also on formalin material. Since formalin is always available at postmortem examinations and permits the use of so many differential stains, it would seem to be the best fixation for purposes of routine examination.

For certain animals and for specific purposes some highly differential techniques have recently been devised by Cleveland and Wolfe [19] and Severinghaus.[20] Gathering material for this study commenced

thirteen years ago, and in order to make results comparable the same method has been followed throughout. These newer methods have therefore not been used for the quantitative work on this human material. It may not be amiss to call attention also to a paper by Weiss,[21] recently published, which further broadens the application of Mallory's connective tissue stain in general histological technique.

Specimens were taken at autopsy by carefully pinching off the sella turcica with heavy bone forceps, leaving the hypophysis *in situ*, and putting the entire mass immediately in formalin and transporting it to the laboratory where the hypophysis was dissected out.* The dura and another very thin layer of connective tissue, representing the arachnoid membrane, were carefully removed except from about the posterior lobe, where these meninges are so adherent that it is inadvisable to attempt to remove them if the posterior lobe is to be kept intact. Only in cases of sudden and usually accidental death, where there were no evident hypophyseal involvements, was material used.

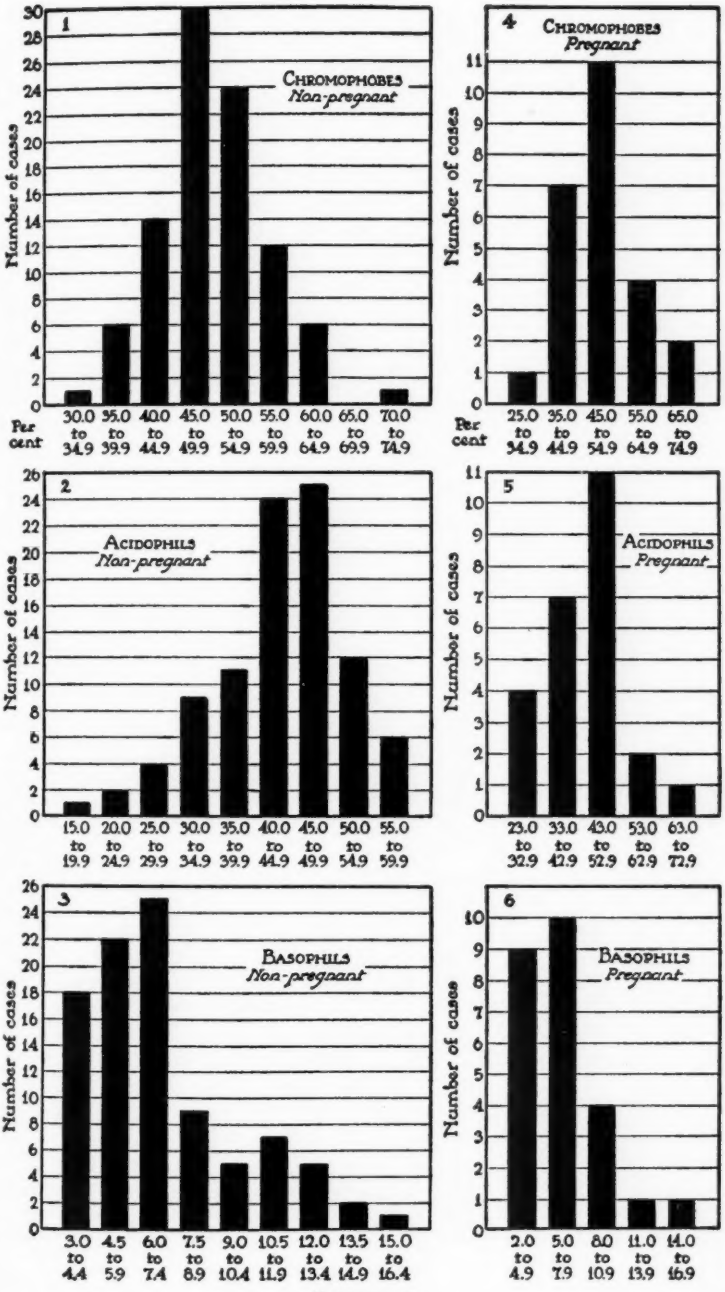
The results have been tabulated by means of the commoner statistical methods, which are necessary for properly evaluating differences, especially where these are relatively small and the number of cases limited. Lack of statistical constants and details of methods used render much published data almost valueless for scientific purposes.

Results

Frequency Distribution: The distribution of the percentages of the three types of cells of 94 non-pregnant and 25 pregnant females is shown graphically in Charts 1 to 6. These graphs and subsequent tables are based on an average of 214 sample fields and an average total, in round numbers, of 15,000 cells in each hypophysis (about 70 cells per field of oil immersion lens). The basophils are distinctly skewed in the positive direction, as is also the case in the male[1]; but otherwise there is the usual tendency to form a fairly symmetrical curve.

Percentage of Chromophobes in Non-Pregnant Females: Table I shows that the chromophobes vary from 33 per cent to 74 per cent and average between 49 and 50 per cent. The coefficient of variation

* For the coöperation of various members of the Department of Pathology of the University of Minnesota, and especially Drs. J. S. McCartney and B. J. Clawson, in securing specimens, the author is greatly indebted.



CHARTS 1–6

Frequency distribution of the percentage of the different types of cells in the anterior lobe of the hypophysis of 94 non-pregnant and 25 pregnant females.

is 14, which is of the same order as the variability of the weight of the normal liver.

Percentage of Acidophils in Non-Pregnant Females: The upper and lower limits (Table I) are about 15 per cent lower than the chromophobes, but the average (about 44 per cent) is only 6 per cent less. The coefficient of variation is 19. This is slightly greater than the variability in the normal weight of the heart.

Percentage of Basophils in Non-Pregnant Females: The basophils (Table I) show a relatively greater individual variation, ranging from 3 to nearly 16 per cent, with an average of 7 per cent. The co-

TABLE I

Percentage of the Different Types of Cells in Anterior Lobe of Human Hypophysis (94 Normal, Non-Pregnant Females)

Cell type	Minimum	Maximum	Median	Mean and prob. error	Standard deviation	Coefficient of variation
Chromophobe . . .	32.9	74.1	49.0	49.6 ± 0.47	6.79	14
Acidophil	19.2	57.5	44.2	43.4 ± 0.56	8.10	19
Basophil	3.0	15.6	6.6	7.0 ± 0.20	2.94	42

efficient of variation 42 means that they are nearly as variable in relative number as the thyroid varies in weight, the corresponding constant for the thyroid gland being 45.

Percentage of the Different Types of Cells with Age: Table II shows that when the 33 cases above 50 years of age (average 61 years) are

TABLE II

Mean Percentage of the Different Types of Cells in the Anterior Lobe of the Hypophysis According to Age (Non-Pregnant Females)

Cell type	50–84 yrs. (33 cases)	16–49 yrs. (61 cases)	Difference	P.E. of difference	Difference / P.E. of difference
Chromophobe . . .	52.2 ± 0.81	48.2 ± 0.54	+4.0	0.97	4.1
Acidophil	39.6 ± 0.93	45.5 ± 0.64	−5.9	1.13	5.2
Basophil	8.1 ± 0.37	6.4 ± 0.22	+1.7	0.43	4.0

compared with those under 50, whose average age is 31 years, the chromophobes are 4 per cent and the basophils nearly 2 per cent higher and the acidophils 6 per cent lower in the older group. The

last column indicates the ratio of the differences to the probable error of these differences. Since in each case this ratio is 4 or more, this is statistically significant. A ratio of 4 means that there is less than one chance in a hundred that the difference is merely due to random sampling. The same was true in the male, except there the increase in the basophils with age was too small to be significant. The prevalent idea one obtains from the literature is the same as indicated in these females.

Relation of Types of Cells to Stature: The average stature of the 94 non-pregnant females was 162.2 cm. (median 162 cm.) and of the 25 pregnant females 162.7 cm. (median 162 cm.). This is very close to the average stature for this region.[22] A comparison of those below the average with those above the average is found in Table III, with a test of the significance of the differences. The slight differences are

TABLE III

Comparison of the Mean Percentages of the Different Types of Cells in Hypophyses of Non-Pregnant Females Below Average Stature with Those Above Average Stature

Cell type	Stature 163–178 cm. (45 cases)	Stature 136–162 cm. (49 cases)	Difference	P.E. of difference	Difference / P.E. of difference
Chromophobe . . .	50.4 ± 0.74	48.8 ± 0.60	+1.6	0.95	1.7
Acidophil	42.3 ± 0.86	44.4 ± 0.73	−2.1	1.13	1.9
Basophil	7.2 ± 0.25	6.8 ± 0.31	+0.4	0.40	1.3

not significant. To be significant the ratios in the last column should be at least 3. In spite of the pathological and experimental evidence for the close association of the acidophils with growth processes, there is no suggestion here along that line, confirming the previous report on the male.[1]

Effect of Pregnancy on the Relative Number of the Different Types of Cells in the Anterior Lobe of the Hypophysis: There is much disagreement in the statements concerning the histological changes found in the hypophysis during pregnancy.[7, 8] A fairly common notion is that there is increased acidophilia, either due to an increase in the number of acidophils or the development of a new cell from chromophobes — the so-called "pregnancy cell" — which in the human is said to be more or less acidophilic in staining properties, so much so that Bailey [23] speaks of it as a *beta* cell. There are, however, others who

regard it as more like a chromophobe.[24] Biedermann,[17] after a special study of the tinctorial reaction of these cells, decided that they are enlarged chromophobes with slight basophilic properties.

With the technique and material here involved and attempts to chromate sections from the formalinized tissue and apply the staining method of Cleveland and Wolfe,[19] Kernohan,[16] and other methods, as well as the hematoxylin and eosin stain, no special cell could be found that was characteristic of the hypophysis of females who had been pregnant only a few days before death or who even had a fetus *in utero* when the postmortem was performed. It is true there were only 4 at, or near full term, and that half of them were less than six weeks pregnant, but the rest of the 25 cases occurred in the

TABLE IV

Percentage of the Different Types of Cells in Anterior Lobe of the Hypophysis of 25 Pregnant Females

Cell type	Minimum	Maximum	Median	Mean and prob. error	Standard deviation	Coefficient of variation
Chromophobe . . .	26.6	71.0	49.6	50.1 ± 1.26	9.33	19
Acidophil	23.3	64.5	45.7	43.4 ± 1.34	9.91	23
Basophil	2.1	14.3	5.7	6.3 ± 0.51	3.04	48

intervening months, so that there should be some evidence of any marked departure from the appearance of the hypophysis in non-pregnant females.

One gains the general impression that possibly the chromophobes have on the average more cytoplasm, as if the smaller ones have hypertrophied to some extent. Extensive measurements of the size of cells would have to be made to verify this, or show conclusively any other definite change in the size of any of the types of cells. Unusually large chromophobes, different from anything that could be found in the hypophysis of non-pregnant females, were not found. While the cell cords in some pregnant females appeared slightly larger, this was not marked enough to constitute a criterion by which the hypophyses of pregnant females could be distinguished from those of non-pregnant ones.

The results are recorded in Table IV. If the cells were decidedly acidophilic in pregnancy they were counted in with ordinary acido-

phils. On the other hand, if they were like chromophobes tinctorially, they were included with the chromophobes. There seems to be no essential difference between these cases where pregnancy was involved and the non-pregnant ones (Table I). The coefficients of variation are somewhat greater in the group of pregnant females, as might be expected from the small number of cases. How they compare with the figures from a similar age group of 61 non-pregnant females is presented in Table V. There are apparently 2 per cent less acidophils and a corresponding increase in chromophobes in the group of pregnant females, which falls in line with the report of Biedermann[17] and some others, including the recent studies of Wolfe and Cleveland, [24] but the ratio of these differences to the probable error

TABLE V

Comparison of the Mean Percentages of the Different Types of Cells in the Anterior Lobe of the Hypophysis in Pregnant Females and Non-Pregnant Females Under 50 Years of Age

Cell type	Pregnant group 15–39 yrs. (25 cases)	Non-pregnant group 16–49 yrs. (61 cases)	Difference	P.E. of difference	Difference / P.E. of difference
Chromophobe . . .	50.1 ± 1.26	48.2 ± 0.54	+1.9	1.37	1.4
Acidophil	43.4 ± 1.34	45.5 ± 0.64	−2.1	1.48	1.4
Basophil	6.3 ± 0.51	6.4 ± 0.22	−0.1	0.56	0.2

of the differences is only 1.4, and hence the differences are not significant. A comparison of the cases in late stages of pregnancy with those in early stages similarly shows no significant differences, but the number of individuals is too small to warrant any definite statement on this particular point.

From this it is evident that the enlargement of the anterior lobe of the hypophysis during pregnancy (most marked during advanced stages [25] — confirmative quantitative data on these cases will be published later) is not due to hyperplasia of any particular type of cell. The largest hypophyses in cases of pregnancy in our collection (over 1 gm. in weight) appear distinctly edematous, rather than hyperplastic.

Sex Differences in Cell Proportions in the Anterior Lobe of Adult Human Hypophysis: In Table VI are shown the averages of 100 supposedly normal males (from previous publication [1]) and of the 94

non-pregnant females, with a test of the significance of the differences. There are nearly 3 per cent less chromophobes and 4 per cent less basophils in the female than in the male, with a corresponding (6 to 7 per cent) larger number of acidophils in females. These differences are strictly significant, as judged by the ratios in the last column of Table VI.

TABLE VI

Comparison of the Mean Percentages of the Different Types of Cells in the Anterior Lobe of the Hypophysis in Males and Non-Pregnant Females

Cell type	Males (100 cases)	Non-pregnant females (94 cases)	Difference	P.E. of difference	Difference / P.E. of difference
Chromophobe . . .	52.2 ± 0.54	49.6 ± 0.47	+2.6	0.72	3.6
Acidophil	36.8 ± 0.52	43.4 ± 0.56	−6.6	0.76	8.7
Basophil	10.9 ± 0.25	7.0 ± 0.20	+3.9	0.32	12.2

If only the cases under 50 years of age are considered, in order to eliminate the changes incident to old age and menopause, the sex differences are equally as marked and statistically significant (Table VII).

Since we have just shown that there is, if anything, a decrease in the proportion of the acidophils during pregnancy, the higher per-

TABLE VII

Comparison of the Mean Percentages of the Different Types of Cells in the Anterior Lobe of the Hypophysis in Males and Non-Pregnant Females Under 50 Years of Age

Cell type	Males 18–50 yrs. (69 cases)	Non-pregnant females 16–49 yrs. (61 cases)	Difference	P.E. of difference	Difference / P.E. of difference
Chromophobe . . .	51.1 ± 0.56	48.2 ± 0.54	+2.9	0.78	3.7
Acidophil	38.1 ± 0.63	45.5 ± 0.64	−7.4	0.90	8.2
Basophil	10.8 ± 0.31	6.4 ± 0.22	+4.4	0.38	11.6

centage of acidophils in females as compared with males is evidently not due to the factor of pregnancy having been involved at some time or another. Naturally, the next step would be to see at what age period this sex difference arises. Hypophyses from children are much more difficult to obtain, so that unless some coöperative effort is made, this point will remain a question for a considerable time.

Summary

1. The results are given of a determination of the relative number of chromophobes, acidophils (*alpha* cells) and basophils (*beta* cells) in the anterior lobe of the hypophysis of 94 carefully selected and supposedly normal, formalin-fixed hypophyses of non-pregnant females 16 to 84 years of age (average 42 years) from cases of sudden or accidental death, and of 25 pregnant females 15 to 39 years of age (average 27 years).

2. The data were obtained by counting all the cells containing nuclei in an average of 214 equally spaced microscopic fields from three different, well separated sections 5 microns in thickness from each hypophysis, and by using Mallory's connective tissue stain after slight staining with hematoxylin to bring out the nucleus.

3. While the above constitutes a highly differential stain that is very easily applied, attention is called to an increasing variety of methods that are being used in an effort to differentiate sharply between the types of cells in the anterior lobe of the hypophysis.

4. In the group of non-pregnant females the chromophobes average between 49 and 50 per cent of all the cells, with a coefficient of variation of 14; the acidophils average between 43 and 44 per cent, with a coefficient of variation of 19; and the basophils average 7 per cent, with a coefficient of variation of 42. These figures are radically different from those given for a mixed group of females and a group of female dementia praecox cases by McCartney,[26] in both of which he reports three times as many basophils and only half as many chromophobes. How much of these differences is pathological, and how much is due to the technique used, cannot be determined for lack of details.

5. Female above 50 years of age (average 61 years) show on an average 4 per cent more chromophobes, nearly 2 per cent more basophils and 6 per cent fewer acidophils than those below 50 years of age (average 31 years).

6. There is no correlation between body length and any particular type of cell.

7. While there are on an average relatively fewer acidophils and more chromophobes in the pregnant females than in the non-pregnant, the differences are too small to be statistically significant.

8. Contrary to the opinion of many, the enlargement of the

anterior lobe of the hypophysis during pregnancy is apparently not due to marked hyperplasia of any one of the three generally recognized types of cells, nor could a special so-called "pregnancy cell" be identified.

9. Females have a distinctly higher proportion of acidophils than males, and males, on the other hand, have a higher percentage of chromophobes and basophils.

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THE RELATION OF HEPATITIS TO CHOLECYSTITIS *

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The relation of inflammatory lesions of the liver to cholecystitis has been a question for many years. The clinical observations of Riedel [1] who noted an enlargement of the right lobe of the liver in cases of chronic cholecystitis must be considered one of the earliest references to the association of these two conditions. Charrin and Roger [2] first called attention to the fact that the injection of bacteria into the common bile duct produced a cholecystitis that was associated with suppurative lesions of the liver. Later Quincke [3] and others also mentioned enlargement of the liver in cholecystitis. It was not, however, until Graham [4] in 1918 published his studies on biopsies of the liver taken at operation in 30 cases of cholecystitis that the conception of hepatitis as a source of gall-bladder infection received much consideration. Since that time many surgical authorities, both in this country and abroad, have accepted Graham's dictum that hepatitis constantly accompanies and is frequently the source of infection in cholecystitis. Graham described an enlargement of the liver and other gross changes such as adhesions and rounding of the liver edge in 87 per cent of his series of cases. Later, however, he [5] stated: "We are forced to the conclusion that the actual percentage of noticeable enlargement of the liver is very much smaller than the figure given." He felt that the increase in size was due to edema, and in the acute and subacute cases he noted microscopically the constant presence of a hepatitis consisting of an infiltration of lymphocytes and polymorphonuclear leukocytes in the interlobular connective tissue around the bile ducts and vessels. Mild degrees of fatty metamorphosis and other minor changes in the parenchyma of the liver were also noted. In the cases of chronic cholecystitis he described a picture that resembled biliary cirrhosis and suggested the hepatitis of the acute and subacute cases as a possible etiological factor in the production of this condition. In describing these chronic cases, how-

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ever, he does not mention the presence or absence of biliary obstruction, which is conceded by certain workers — namely Charcot and Gombault [6] and others — to produce a picture of cirrhosis in experimental animals. McCartney [7] and MacMahon and Mallory [8] have shown a similar condition in man following prolonged obstruction of the common or hepatic bile ducts. The biopsies of the liver that Graham [4] removed from his surgical cases were taken from the edge of the liver, which is an extremely unreliable area from which to judge the condition of the liver as a whole. The picture of cirrhosis is frequently seen in this region even in normal livers.

Three years after Graham's original contribution Peterman, Priest and Graham [9] postulated a lymphatic theory for the etiology of cholecystitis. Basing this theory on the work of Sudler,[10] who showed an intimate connection between the lymphatics of the gall-bladder and extra- and intrahepatic lymphatics, they expressed the opinion that, at least in a certain percentage of cases, infection reached the liver through the portal blood where it produced an inflammatory lesion, and that the infection was conducted from the liver by the lymphatics to the gall-bladder. They quoted Mall's work [11] on the lymph channels of the liver, pointing to the fact that since the only demonstrable lymph vessels of the liver substance were situated in the portal spaces, this area would naturally be the first to show evidence of such an infectious process. After the establishment of an infection in the gall-bladder they felt that a retrograde lymphatic infection of the liver from the gall-bladder might keep up a "vicious cycle" that would necessitate the removal of the gall-bladder to effect a clinical cure. In support of this theory they did certain experimental work on dogs. The animals were subjected to preliminary laparotomies and biopsies of the livers were taken and studied. At the same time the cystic ducts and cystic arteries were ligated and the animals were given intravenous injections of 24 hour cultures of non-hemolytic streptococci or *B. coli*. These animals were killed at the end of 19 days and their livers and gall-bladders were studied. The livers that had been normal, as proved by the original biopsies, showed a hepatitis that was most pronounced in the region of the gall-bladder bed. Later Graham and Peterman [12] continued this line of experimentation but varied it by giving one or two injections of bacteria into the portal vein, with much the same results.

Peterman [13] in 1923 reviewed the whole subject of gall-bladder infection, repeated many of the procedures previously done by himself and other experimenters and in addition he injected bacteria into the appendiceal veins of dogs and noted not only the previously mentioned hepatitis but also a pancreatitis. In his book, published some years later, Graham [5] described a similar hepatitis in patients operated upon for chronic appendicitis, but was unable to explain why these patients showed no evidence of gall-bladder infection.

The whole lymphogenous theory of cholecystitis advocated by Graham is based on the work of Sudler.[10] Sudler claimed an intimate lymphatic connection between the liver and gall-bladder. Winkenwerder,[14] however, using more refined technical methods, recently came to the conclusion that in the cat there was no lymphatic connection between these organs and so raised some question as to Graham's original thesis.

Graham's ideas, however, have been widely accepted and his work has received confirmation from many sources. Judd,[15] and Judd and Marshall,[16] Deaver,[17] Moynihan,[18] and Heyd, MacNeal and Killian,[19] using Graham's original methods of investigation have largely confirmed his work. Deaver believed, however, that the original lesion occurred in the gall-bladder and that the liver was secondarily involved. Tietze and Winkler,[20] studying biopsies of the liver from operative cases, took their specimens from the dome of the liver to eliminate errors that might creep in from examination of the liver edge. They also agreed with Graham. Mentzer,[21] in a study of 612 consecutive autopsies, showed that the frequency of hepatitis practically paralleled the occurrence of cholecystitis. Martin,[22] however, without any stated clinical or experimental data reasoned that the inflammatory reactions in the liver described by Graham were probably secondary to tissue irritation resulting from the absorption of bacteria and toxic substances from the gastrointestinal tract. Aiello,[23] in a postmortem study of 43 individuals dead of infectious diseases, found only 6 instances of definite inflammatory changes in the portal spaces of the liver, and in a series of 14 cases of phthisis he found similar but less marked reactions. He believed that biliary tract infection was not a necessary accompaniment of cholecystitis.

The following study was attempted in an effort to determine how frequently the inflammatory lesions of the liver described by Graham

occur in autopsied cases, and what relation, if any, this lesion bears to inflammatory conditions of the gall-bladder. A total of 212 autopsies was studied. These were unselected but not consecutive. At first it was thought desirable to exclude the autopsies on newborn infants and young children, but as the work developed it became evident that a study of all age groups was indicated, so that 15 cases under 1 year of age and 10 cases between 1 and 10 years of age were included. The work was carried out in the following manner: From each case three blocks of liver and at least one of the gall-bladder were removed. In most instances the tissue from the gall-bladder was taken from the portion of the organ covered by peritoneum, but in a few cases pieces of the gall-bladder were taken through the gall-bladder bed. One section of the liver was always taken from the free margin of the organ, just to the right of the gall-bladder. Two other blocks were removed from the substance of the right lobe well away from the capsule. The tissue was fixed in 10 per cent formalin and stained with hematoxylin and eosin. The gross appearance of the liver and gall-bladder was noted. Only those gross changes of the gall-bladder that were beyond question were recorded in order to simplify tabulation and to exclude non-inflammatory lesions from the group showing cholecystitis. Cholesterosis was not recorded. In the gross descriptions of the liver such changes as chronic passive congestion, increase in fat content, alterations of the capsule and the presence of tumor masses or abscesses were noted. The cases were studied with reference to age and sex, and the cause of death was recorded in each case. The liver and gall-bladder were studied microscopically.

Regardless of gross findings the decision as to whether or not the lesions of the gall-bladder should be called cholecystitis rested on the microscopic examination. Only those gall-bladders showing an infiltration of lymphocytes or leukocytes or both were called cholecystitis. The degree of inflammatory change in the gall-bladder wall was graded as 1, 2, 3 and 4. In Figures 1, 2, 3 and 4 photomicrographs of representative cases in each of the several grades are shown. In the microscopic study of the liver sections three observations are recorded: (1) The degree of infiltration in the portal spaces. This was arbitrarily divided into 4 grades indicated as 1, 2, 3 and 4, and examples of each grade are shown in Figures 5, 6, 7 and 8. (2) The presence or absence of polymorphonuclear leukocytes

was charted and some estimate of their number was attempted by indicating them as "many" or "few." This fact was noted because, especially in the less marked grades of infiltration, the presence of lymphocytes alone is said by some histologists (Maximow and Bloom[24]) to be a normal occurrence. (3) Varying degrees of cirrhosis were also noted. Most of these were the non-clinical or latent type.

As mentioned above, Graham's[4, 5] description of hepatitis emphasized particularly periportal infiltration and this lesion alone was studied in this series. Parenchymal changes were disregarded because the variety of pathological lesions causing death produced changes in the liver that had no possible relation to the hepatitis. Tietze and Winkler,[20] in their description of hepatitis, stressed changes in the bile ducts such as desquamation of epithelium and the presence of leukocytes in the lumen of the ducts. These changes were not noted by Graham and they were not seen in this series.

The cases were first divided into 10 age groups. Those cases below 1 year of age were placed in one group. The balance of the cases were grouped by decades. All those over 80 years of age were, however, considered together.

In the age group under 1 year the newborn infants were considered separately and the findings are shown in the first portion of Table I. Here, in all cases, a certain amount of infiltration was noted in the portal spaces of the liver. This reaction can hardly be considered as inflammatory in character and a possible explanation is offered by the fact that in all but 1 case myelocytes were seen in the infiltration. The hemopoietic function of the liver is generally considered to be located entirely in the blood islands that are located in the liver lobule. This fact is recognized by Maximow and Bloom,[24] and by Downey.[25] Nevertheless, the presence of myelocytes in this region indicates the possibility that the portal areas of the liver may also have a hemopoietic function in the newborn.

Most of the remaining cases in this first group, namely those living a period of several months, showed a minimal amount of infiltration in the portal spaces. In the group as a whole 2 Grade 3 reactions were noted, 1 occurring in a newborn infant and the other in an infant dying of primary peritonitis. In the first instance no explanation is offered but in the second case the infiltration was possibly secondary to a general sepsis.

The findings in the gall-bladders of this group were also interest-

ing. In 6 of the cases, both polymorphonuclear leukocytes and lymphocytes were seen, while in 3 others lymphocytes alone were noted.

TABLE I

Cases Under 1 Year of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver				Gall-bladder		Cause of death
					Portal infiltration	Myelocytes	Polymorphonuclears	Connective tissue	Leukocytes	Lymphocytes	
343	1 hr.	M	–	–	1	+	Many	–	o	o	Congenital absence of rectum
344	11 hr.	M	–	–	1	–	Many	–	+	..	Asphyxia
9	N. B.	F	–	–	1	+	Many	–	+	..	Birth trauma
10	N. B.	M	–	–	1	+	Many	–	..	+	Birth trauma
75	N. B.	M	–	–	2	+	Many	–	–	–	Prematurity, birth trauma
40	2 hrs.	M	–	–	2	+	Many	–	..	+	Prematurity
425	N. B.	M	–	–	3	+	Many	–	+	..	Birth trauma
7	3 mos.	M	–	–	1	–	Many	–	o	o	Bronchopneumonia
11	6 mos.	F	–	–	1	–	Few	–	..	+	Meningitis, otitis media
77	5 mos.	F	–	–	1	–	Many	–	–	–	Bronchopneumonia
87	1 mo.	M	–	–	1	–	Many	–	–	–	Inanition
96	6 mos.	M	–	–	1	–	Many	–	+	..	Otitis media, cervical cellulitis
67	4 mos.	M	–	–	1	–	Few	–	o	o	Bronchopneumonia, empyema
66	2 mos.	M	–	–	–	–	Few	–	+	..	Bronchopneumonia
43	1 mo.	M	–	–	3	–	Many	–	+	..	Peritonitis, bronchopneumonia

In all tables ± = minimal infiltration; + = presence of cells indicated; – = absence of cells indicated; o = no observation; 1, 2, 3, and 4 = degree of portal infiltration or cholecystitis.

N.B. = new born; C.P.C. = chronic passive congestion; Tbc. tuberculosis; Carc. = carcinoma.

These cells, however, did not occur in sufficient numbers to indicate an inflammatory process and lymphocytes are known to occur

normally in the gall-bladders of infants. With these findings, it is interesting to speculate as to whether or not many of the minimal cellular reactions, seen in the gall-bladders of adults removed surgically, really represent a pathological process.

In the 1 to 10 age group (Table II) the degree of periportal infiltration was less marked than in any other single group. In 1 case there was no portal infiltration and in 3 others there was only a minimal degree. Only 1 case showed any evidence of cholecystitis. This gall-bladder showed a definite inflammatory change that was unquestionably of clinical significance. The patient died as the

TABLE II

Cases From 1 to 10 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
42	3	M	–	–	–	–	–	–	Mastoiditis
329	4	F	–	–	±	Few	–	–	Skull fracture, bronchopneumonia
354	5	M	–	–	±	Few	–	–	Skull fracture
407	3	M	–	–	±	Few	–	–	Diphtheria (laryngeal)
370	3	M	–	–	1	Few	–	–	Appendicitis, peritonitis
65	1	M	–	–	1	Many	–	–	Ulcerative colitis
99	2	F	Tbc.	–	1	Few	–	–	Pulmonary tuberculosis
332	8	M	Torn	–	2	Few	–	–	Ruptured liver
418	8	M	–	–	3	Many	–	3	Skull fracture
100	3	F	–	–	3	Many	–	–	Scarlet fever

result of a skull fracture and no clinical history was available relative to gall-bladder complaints.

In the next 2 groups, namely the 10 to 20 and 20 to 30 age groups (Tables III and IV), the finding of a periportal infiltration in varying degrees was a constant factor, and sometimes it was quite pronounced. In these younger age groups it is interesting to note the high percentage of cholecystitis that was found. Eusterman,[26] Snyder [27] and others have emphasized the fact that cholecystitis is more frequent in young individuals than is usually recognized. Eusterman reviewed a series of cases seen in the Mayo Clinic and

TABLE III

Cases From 10 to 20 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
401	19	M	Abscesses	Thick	1	Few	–	1	Abscesses of liver, peritonitis
437	11	F	–	–	1	Few	–	0	Suppurative meningitis
14	12	M	–	–	1	Few	–	–	Skull fracture
21	18	F	C. P. C.	–	1	Few	–	1	Bacterial endocarditis
319	19	M	–	Opaque	2	Many	–	1	Septic sore throat, empyema
389	16	M	–	Opaque	2	Many	–	–	Pulmonary tuberculosis
423	13	F	Fibrin	–	2	Many	–	–	Appendicitis peritonitis
355	19	M	Abscess	Opaque	3	Many	–	3	Abscesses of liver, appendicitis

TABLE IV

Cases From 20 to 30 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
336	26	F	–	–	1	Few	–	1	Ruptured uterus
339	27	F	Fatty	–	1	Few	–	0	Pulmonary tuberculosis
432	29	M	C. P. C.	Thick	1	Few	–	–	Rheumatic endocarditis
38	20	F	C. P. C.	Thick	1	Few	–	3	Congenital heart, bacterial endocarditis
70	29	M	–	–	1	Many	–	–	Pulmonary tuberculosis
91	21	F	–	Opaque	1	Few	–	–	Tuberculosis, endocarditis
82	23	F	–	–	2	Few	–	1	Lobar pneumonia
23	21	F	–	–	2	Many	–	–	Suppurative meningitis
19	23	F	Scar	Thick	2	Few	–	2	Hypertensive kidneys
415	29	M	–	Thick	2	Many	–	1	Lobar pneumonia
361	25	M	–	Thick	2	Few	–	1	Skull fracture, ruptured liver
413	21	M	–	–	3	Many	–	–	Bacterial endocarditis
414	27	M	–	–	3	Many	–	1	Amyloid disease
420	28	F	–	–	3	Few	–	3	Abortion, peritonitis
36	27	F	–	Stones	3	Few	–	1	Mastoiditis, meningitis
45	21	M	–	–	3	Few	–	–	Frozen feet, pneumonia
362	25	F	–	–	4	Few	+	2	Pulmonary tuberculosis

concluded that in males under 25 and females under 20 years of age, clinical gall-bladder disease occurred in about 0.85 per cent of cases. The cases were about equally distributed between the sexes. He pointed out the association of this condition with non-specific infectious processes such as appendicitis, tonsillitis and rheumatic fever. Graham predicted that our more modern methods of diagnosis would soon show a considerably higher percentage of cases of cholecystitis in children than is generally recognized.

TABLE V

Cases From 30 to 40 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
326	30	M	–	–	±	Few	–	–	Epilepsy, pneumonia
356	30	M	–	–	1	Few	–	–	Poliomyelitis
396	39	F	C. P. C.	Opaque	1	Few	–	–	Valve defect
405	36	F	–	–	1	Few	–	–	Diabetes, pneumonia
15	37	F	–	–	1	Few	–	–	Ruptured ovarian cyst
20	35	M	Scar	–	1	Few	–	1	Pulmonary tuberculosis
85	30	F	–	–	1	Few	–	–	Alcoholism, fatty liver
347	36	F	–	–	2	Few	–	0	Ruptured gut, peritonitis
352	37	M	Scar	–	2	Many	–	1	Gunshot wound, hemorrhage
369	37	M	Scar	–	2	Many	–	1	Lobar pneumonia
391	38	M	–	–	2	Many	–	1	Appendicitis, peritonitis
13	35	M	–	–	2	Few	+	–	Skull fracture, pneumonia
24	31	F	–	–	2	Few	–	–	Bronchopneumonia
84	33	F	–	Thick	2	Few	–	2	Bronchiectasis, pneumonia
98	37	M	–	–	2	Few	–	–	Bronchiectasis, pneumonia
71	37	F	Rough	Thick	4	Many	+	3	Cirrhosis of the liver

In the various age groups above 30 years a certain degree of portal infiltration was noted in all cases. The definite gross findings in both liver and gall-bladder showed an irregular, progressive increase with age, but the cases showing cholecystitis microscopically showed no constant relation to the degree of portal infiltration or to the gross changes incident to age. Tables V, VI, VII, VIII, IX and X tabulate the findings in these groups. Here, also, appeared certain cases of clinical and latent cirrhosis.

In the younger age groups there seemed to be some suggestion that in cases of generalized infection the infiltration in the portal spaces was more marked, but in the older groups this association

TABLE VI

Cases From 40 to 50 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
325	42	M	Fatty	–	1	Many	–	0	Skull fracture
382	46	M	–	Thick	1	Few	–	2	Cerebral hemorrhage
435	41	F	Scar	Thick	1	Few	–	2	Valve defect, pneumonia
2	43	M	C. P. C.	–	1	Many	–	–	Skull fracture, meningitis
6	44	M	–	–	1	Few	–	–	Pulmonary tuberculosis
16	47	F	Scar	–	1	Few	–	–	Huntington's chorea
82	46	M	–	–	1	Many	–	–	Bronchopneumonia
89	49	F	–	–	1	Few	–	–	Carcinoma of the ovary
95	49	F	–	–	1	Few	–	–	Pulmonary tuberculosis
1202	40	M	–	–	1	Few	–	2	Ruptured kidney, pneumonia
2	48	F	–	–	1	Few	–	–	Hypertensive kidney
6	40	M	–	–	1	Few	–	–	Suppurative meningitis
313	42	M	Scar	–	2	Many	–	–	Pulmonary tuberculosis
323	45	F	–	Thick	2	Many	–	–	Bronchopneumonia
348	47	M	–	–	2	Few	–	–	Chronic glomerulonephritis
366	42	M	Fatty	–	2	Few	–	1	Chronic alcoholism, pneumonia
372	40	M	Torn	Stones	2	Few	–	2	Ruptured liver
392	48	M	–	–	2	Many	–	–	Pyonephrosis
29	44	M	–	Opaque	2	Few	–	1	Coronary thrombosis
61	49	M	–	–	2	Few	–	–	Skull fracture, meningitis
101	45	F	–	Opaque	2	Few	–	–	Otitis media, meningitis
1385	40	M	Scar	–	2	Many	–	–	Fractured spine
15	42	M	–	–	2	Many	–	1	Pulmonary tuberculosis
17	45	F	C. P. C.	Thick	2	Many	–	2	Bacterial endocarditis
20	44	M	–	–	2	Few	–	1	Fractured spine, myelitis
365	40	M	–	–	3	Many	–	–	Gunshot wound
49	47	M	–	–	4	Many	–	2	Carcinoma of stomach
342	41	M	–	–	4	Few	–	1	Undetermined, poison (?)

was not at all evident. With the idea of discovering whether or not the presence in the gastro-intestinal tract of a chronic ulcerative lesion influenced the degree of infiltration in the liver, those cases showing this type of lesion were tabulated separately (Table XI).

TABLE VII

Cases From 50 to 60 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
327	59	M	–	–	1	Few	–	–	Carcinoma of pharynx
364	51	F	Torn	–	1	Few	–	1	Skull fracture
387	52	M	–	Thick	1	Few	–	1	Hypertensive heart
408	52	M	–	Thick	1	Few	–	2	Hypertensive heart
441	50	M	Scar	Thick	1	Few	–	2	Skull fracture
4	59	F	Scar	–	1	Few	–	–	Cerebral malacia, pneumonia
54	53	M	–	–	1	Few	–	–	Cerebral hemorrhage
73	55	F	Tumor	–	1	Few	–	–	Carcinoma of the breast
368	50	F	Scar	–	2	Few	–	–	Bronchiectasis, pneumonia
404	53	F	–	–	2	Many	–	–	Pulmonary embolus
428	58	M	–	Thick	2	Few	–	Carc.	Bronchiectasis, pneumonia
442	50	M	–	–	2	Few	–	–	Skull fracture
444	50	M	–	–	2	Few	–	–	Skull fracture
28	50	M	Scar	Stones	2	Many	+	–	Skull fracture, endocarditis
31	50	M	–	–	2	Few	–	–	Intestinal obstruction
34	52	F	–	Stones	2	Many	–	–	Hypernephroma, pneumonia
74	51	F	–	Stones	2	Few	–	–	Valve defect, cerebral malacia
88	52	M	–	Thick	2	Few	–	1	Carcinoma of the testicle
93	53	M	–	–	2	Few	–	1	Cyanide poisoning
10	52	F	Scar	Stones	2	Few	–	1	Cerebral malacia
90	50	M	C. P. C.	Opaque	2	Few	–	–	Lobar pneumonia, cerebral malacia
349	55	M	Scar	–	3	Many	+	–	Lobar pneumonia
381	53	M	–	–	3	Many	–	1	Perforated gastric ulcer
394	55	M	–	Thick	3	Few	–	2	Luetic aortitis, pneumonia
47	59	F	Tumor	Thick	3	Few	–	1	Carcinoma of breast, pneumonia
341	50	F	Rough	–	4	Few	+	–	Cirrhosis of the liver
22	50	M	Rough	Opaque	4	Many	+	2	Cirrhosis of the liver
33	52	M	Rough	–	4	Many	+	–	Mastoiditis, meningitis
88	55	M	–	Thick	4	Many	+	1	Hypertension, pneumonia

It seemed reasonable to think that if the infiltration was secondary to infection reaching the liver from the gastro-intestinal tract through the portal system, in the presence of a chronic ulcerative

TABLE VIII

Cases From 60 to 70 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-gladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
322	64	M	Scar	Thick	1	Few	–	–	Gangrene of leg, infarcts of lung
328	60	M	–	Opaque	1	Many	–	–	Pulmonary tuberculosis
335	61	M	Scar	Opaque	1	Few	–	–	Skull fracture
410	60	M	–	–	1	Few	–	2	Valve defect
419	60	M	–	Thick	1	Few	–	2	Hypertensive heart, pneumonia
439	67	M	–	–	1	Few	–	–	Carcinoma of prostate, pneumonia
63	67	M	–	–	1	Few	–	–	Lobar pneumonia
64	67	M	–	Opaque	1	Few	–	–	Peritonitis, pneumonia
80	68	M	Scar	Stones	1	many	–	1	Carcinoma of prostate, pyelonephritis
277	60	M	–	–	1	Few	–	–	Carcinoma of bladder, pyelonephritis
330	68	M	Rough	Opaque	2	Many	–	–	Hypertension, uremia
334	69	F	Tumor	Stones	2	Few	–	3	Carcinoma of pancreas
345	68	M	–	Stones	2	Few	–	3	Hypertension, pulmonary infarcts
350	65	M	Scar	–	2	Few	–	1	Cerebral hypertension, pneumonia
363	66	M	–	Stones	2	Few	–	–	Lobar pneumonia
376	69	F	C. P. C.	Stones	2	Many	–	4	Coronary sclerosis
380	66	M	–	–	2	Many	–	0	Carcinoma of esophagus, pneumonia
409	61	M	–	Thick	2	Few	–	2	Hypertension, cerebral malacia
443	63	M	–	Thick	2	Many	–	2	Pernicious anemia
26	64	M	Scar	–	2	Few	–	–	Hypertension, broncho-pneumonia
32	62	F	–	–	2	Few	–	–	Stokes-Adams syndrome
46	68	M	–	Stones	2	Few	–	2	Carcinoma of prostate, pneumonia
50	67	F	Scar	–	2	Many	–	1	Multiple sclerosis, pyonephrosis
51	68	M	–	–	2	Few	–	–	Hypernephroma, erysipelas
57	63	F	–	Stones	2	Few	–	–	Bronchiectasis, pneumonia
83	63	F	Scar	Stones	2	Many	–	1	Valve defect
19	60	F	Scar	–	2	Few	–	–	Diabetes, pericarditis
72	61	M	Rough	Opaque	2	Many	+	2	Valve defect
333	60	M	–	–	3	Many	–	0	Pulmonary tuberculosis
338	61	M	–	Fibrin	3	Few	–	1	Tuberculous peritonitis

TABLE VIII (*Continued*)

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
357	67	F	Tumor	Stones	3	Many	+	Carc.	Carcinoma of the gall-bladder
390	62	M	Tumor	Thick	3	Few	–	–	Carcinoma of the esophagus
424	64	F	–	Stones	3	Many	–	Tumor	Carcinoma of the stomach
438	62	M	Tumor	Thick	3	Few	–	1	Carcinoma of colon, peritonitis
48	65	M	–	Stones	3	Many	–	3	Subdiaphragmatic abscess
314	62	M	Rough	Opaque	4	Few	+	4	Carcinoma of colon
324	60	F	Rough	Opaque	4	Few	+	2	Cirrhosis of liver, valve defect
353	64	F	Rough	Stones	4	Many	+	3	Cirrhosis of liver, peritonitis
371	61	M	Scar	Stones	4	Many	–	1	Bronchopneumonia
78	66	M	Abscess	Stones	4	Many	–	3	Appendicitis, peritonitis

lesion the intestinal filter would be damaged sufficiently to allow more than a normal number of bacteria to reach the liver and this increase would be manifested by an increase in portal space infiltration.

It was found that Grade 1 infiltration occurred in 33.3 per cent of the cases showing chronic gastro-intestinal ulcers, Grade 2 in 37.5 per cent, Grade 3 in 20.8 per cent and Grade 4 in 8.3 per cent. In the entire series, Grade 1 infiltration occurred in 36.7 per cent of the cases, Grade 2 in 40.5 per cent, Grade 3 in 13.6 per cent and Grade 4 in 6.1 per cent. While there is a slight increase in the percentage of the higher grades of infiltration, it is doubtful if the increase is sufficient to be significant in such a small group of cases.

Clinical cholecystitis is generally considered to occur in the ratio of about two females to one male. In this series of autopsy cases, however, the percentage of males showing cholecystitis was 41.9 and the percentage of females 46.2 (Table XII). This ratio is roughly in agreement with Mentzer [21] who reported 57 per cent of males and 64 per cent females with cholecystitis in a series of 612 autop-

TABLE IX

Cases From 70 to 80 Years of Age

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
321	77	F	C. P. C.	–	1	Few	–	–	Bronchiectasis, pneumonia
337	70	M	Scar	–	1	Few	–	–	Pulmonary infarcts, gangrene of leg
358	79	F	Scar	–	1	Few	–	–	Hypertension
377	78	M	–	Opaque	1	Few	–	2	Hypertension
386	73	M	–	Opaque	1	Few	–	–	Bronchopneumonia
397	70	F	Scar	–	1	Few	–	–	Hypertension
407	76	M	Scar	–	1	Few	–	2	Fractured ribs, pneumonia
430	73	M	Scar	–	1	Few	–	–	Hypertension, pneumonia
431	74	M	Scar	–	1	Many	–	–	Fractured pelvis
39	74	M	–	–	1	Few	–	1	Gastric ulcer, pneumonia
44	71	M	–	–	1	Many	–	–	Hypertrophied prostate, pyelonephritis
62	73	F	–	–	1	Few	–	1	Bronchopneumonia
8	77	M	Scar	–	1	Few	–	–	Fractured spine, cerebral hemorrhage
83	71	M	–	Opaque	1	Few	–	1	Lobar pneumonia
315	70	F	Scar	–	2	Few	–	–	First and second degree burns
317	73	M	Scar	–	2	Many	–	2	Pyelonephritis, valve defect
340	77	M	C. P. C.	–	2	Many	–	–	Hypertension, abscesses of kidney
359	75	F	–	–	2	Many	–	–	Fractured femur, gangrenous cystitis
378	72	M	–	–	2	Few	–	0	Hypernephroma, pneumonia
383	73	M	–	–	2	Many	–	–	Skull fracture
398	73	M	C. P. C.	Opaque	2	Few	–	–	Coronary sclerosis
399	71	M	–	Thick	2	Many	–	1	Prostatic abscess, peritonitis
422	76	F	–	–	2	Many	–	1	Carcinoma of stomach, peritonitis
426	77	M	Rough	Stones	2	Few	–	–	Carcinoma of stomach, pneumonia
429	72	M	Rough	–	2	Few	+	0	Cerebral hemorrhage
48	73	M	Scar	–	2	Few	–	–	Bronchopneumonia
92	73	F	–	Opaque	2	Few	–	1	Fractured femur, pneumonia
102	77	F	–	Stones	2	Many	–	1	Diabetes, pneumonia
109	73	M	–	–	2	Few	–	–	Valve defect
85	71	F	–	–	2	Few	–	–	Hemorrhage from gastric ulcer
427	77	F	–	Stones	3	Many	–	2	Perforated gastric ulcer, peritonitis
440	77	F	Rough	–	3	Few	–	1	Valve defect, pneumonia
447	76	M	–	Stones	3	Many	+	–	Lobar pneumonia
5	73	F	Scar	–	3	Few	–	1	Lobar pneumonia
94	74	M	Scar	–	3	Many	–	–	Fractured skull, peritonitis

sies, his higher percentages being due to the fact that he included cholesterosis in his cholecystitis group.

The age distribution in the cases showing cholecystitis is shown in Table XIII. The highest percentage of cholecystitis is seen in the 10 to 20, 20 to 30 and 60–70 age groups. It is doubtful, how-

TABLE X

Cases 80 Years of Age and Over

Number of case	Age	Sex	Gross liver	Gross gall-bladder	Microscopic liver			Microscopic gall-bladder	Cause of death
					Portal infiltration	Polymorphonuclears	Connective tissue		
	yrs.								
374	85	F	Scar	Stones	1	Few	–	1	Hypertensive heart
446	85	F	Scar	–	1	Few	–	–	Gangrenous cystitis, pneumonia
17	84	F	–	Stones	1	Few	–	3	Rheumatic endocarditis, pneumonia
25	87	F	Scar	–	1	Few	–	2	Rheumatic endocarditis, pneumonia
384	93	M	–	–	1	Few	–	–	Coronary sclerosis
351	83	M	–	Stones	2	Few	–	–	Fractured spine
379	81	M	Rough	Stones	2	Few	–	2	Hypertensive kidney, pneumonia
412	83	M	–	–	2	Few	–	–	Carcinoma of stomach, peritonitis
445	81	F	–	–	2	Few	–	–	Fractured femur, pneumonia
12	82	M	–	Opaque	2	Few	–	–	Fractured tibia, pneumonia
69	82	F	–	–	2	Few	–	1	Lobar pneumonia, coronary sclerosis
360	102	M	–	–	2	Few	+	–	Hypertrophied prostate, pneumonia
421	85	M	Scar	–	3	Few	–	–	Hypertrophied prostate
436	89	F	–	Stones	3	Many	–	–	Bronchopneumonia

ever, if the number of cases in several of the decades is sufficient to draw conclusions.

In Table XIV the cases where cholecystitis was present are compared with the cases showing normal gall-bladders with reference to the degree of periportal infiltration. While there is a gradual increase in the percentage of cholecystitis, which roughly parallels

the degree of portal infiltration, many cases with grade 2 and 3 portal infiltration show normal gall-bladders.

In studying the original arrangement of cases by decades, the impression is gained that portal infiltration becomes more severe

TABLE XI

Findings in Cases Showing Chronic Ulcerative Lesions of the Gastro-Intestinal Tract

Number of case	Age	Portal infiltration	Microscopic gall-bladder	Intestinal lesions	Cause of death
	yrs.				
438	62	3	1	Carcinoma of colon	Carcinoma of colon, peritonitis
314	62	4	4	Carcinoma of colon	Carcinoma of colon
49	47	4	2	Carcinoma of stomach	Carcinoma of stomach, pneumonia
424	64	3	–	Carcinoma of stomach	Carcinoma of stomach
422	76	2	1	Carcinoma of stomach	Carcinoma of stomach, peritonitis
426	77	2	–	Carcinoma of stomach	Carcinoma of stomach, pneumonia
412	83	2	–	Carcinoma of stomach	Carcinoma of stomach, peritonitis
389	16	2	–	Tbc. enteritis and colitis	Pulmonary tuberculosis
70	29	1	–	Tbc. enteritis and colitis	Pulmonary tuberculosis
91	21	1	–	Tuberculous colitis	Pulmonary tuberculosis
95	49	1	–	Tbc. enteritis and colitis	Pulmonary tuberculosis
15	37	1	–	Tbc. enteritis and colitis	Pulmonary tuberculosis
92	73	2	1	Ulcerative colitis	Fractured femur, pneumonia
29	44	2	1	Ulcerative colitis	Hypertension, coronary thrombosis
381	53	3	1	Gastric ulcer (ruptured)	Perforated gastric ulcer, peritonitis
401	19	1	1	Gastric ulcer (ruptured)	Abscesses of liver, peritonitis
330	68	2	–	Gastric ulcer	Hypertension, uremia
337	70	1	–	Gastric ulcer	Hypertension, pneumonia
427	77	3	2	Gastric ulcer	Perforated gastric ulcer, peritonitis
39	74	1	1	Gastric ulcer	Pneumonia
85	71	2	–	Gastric ulcer	Hemorrhage from gastric ulcer
440	77	3	1	Duodenal ulcer	Valve defects, pneumonia
374	85	1	1	Duodenal ulcer	Hypertension
379	81	2	2	Duodenal ulcer	Hypertension, uremia, pneumonia

as age increases. In Table XV the percentage of infiltration for each age group and each degree of infiltration are charted. It is evident from these figures, however, that the increase in age is not paralleled by an increase in infiltration in any of the four grades.

TABLE XII

Sex Distribution

Age	Number of cases		Cholecystitis		% Cholecystitis	
	Male	Female	Male	Female	Male	Female
yrs.						
20–30	7	10	3	7		
30–40	8	8	4	2		
40–50	20	8	9	2		
50–60	19	10	9	3		
60–70	29	11	15	6		
70–80	22	13	6	7		
80+	7	7	1	4		
Totals	112	67	47	31	41.9	46.2
Under 1	12	3	0	0		
1–10	7	3	0	1		
10–20	5	3	3	1		
Totals	24	9	3	2	12.5	22.2

TABLE XIII

Age Distribution

Age	Number of cases	Cholecystitis	% Cholecystitis
yrs.			
Under 1	15	0	0
1–10	10	1	10.0
10–20	8	4	50.0
20–30	17	10	58.8
30–40	16	6	37.5
40–50	28	11	39.2
50–60	29	12	41.3
60–70	40	21	52.5
70–80	35	13	37.1
80+	14	5	35.7
Totals	212	83	39.1

TABLE XIV

Comparison of Degree of Portal Infiltration

Liver infiltration	Cholecystitis		No cholecystitis	
	No. cases	Per cent	No. cases	Per cent
None	0	0	2	100
±	0	0	4	100
1	23	29.5	55	70.5
2	35	40.6	51	59.4
3	15	51.7	14	49.3
4	11	84.6	2	15.4

TABLE XV

Age Distribution of Portal Infiltration

Portal infiltration	Age 1–10 yrs.		Age 10–20 yrs.		Age 20–30 yrs.		Age 30–40 yrs.		Age 40–50 yrs.		Age 50–60 yrs.		Age 60–70 yrs.		Age 70–80 yrs.		Age 80 + yrs.	
	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%	No.	%
Neg.	1	10.0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0	0
±	3	30.0	0	0	0	0	1	0	0	0	0	0	0	0	0	0	0	0
1 +	3	30.0	4	50.0	6	35.2	6	37.5	12	42.8	8	27.5	10	25.0	14	40.0	5	35.7
2 +	1	10.0	3	37.5	5	29.4	8	50.0	13	46.4	13	44.8	18	45.0	16	45.7	7	50.0
3 +	2	20.0	1	12.5	5	29.4	0	0	1	3.5	4	13.7	7	17.5	5	14.2	2	14.2
4 +	0	0	0	0	1	5.8	1	6.3	2	7.1	4	13.7	5	12.5	0	0	0	0

Discussion

Study of this series of cases shows conclusively that an infiltration of lymphocytes and polymorphonuclear leukocytes in the portal connective tissue is almost a constant finding in livers from autopsy cases. In this series of 212 cases the portal spaces were free from inflammatory cells in only 2 instances. In 3 other cases the reaction was practically negligible. From these minimal changes the lesion ranged in intensity to the Grade 4 reaction shown in Figure 8. A marked variation in degree was noted in different areas of the same section and only by repeated study of the sections was it possible to arrive at a fair estimate of the intensity of the reactions. As mentioned above, the liver edge showed more intense changes in many cases, both from the standpoint of inflammatory cells and increased connective tissue, than areas well away from the capsule and edge, so that it is justifiable to say that the liver edge is not a reliable source of tissue from which to judge the histology of the whole organ. The published descriptions of the hepatitis accompanying cholecystitis are on the whole rather vague, Tietze and Winkler's [20] description being the only exception, but apparently the basis for all of the accepted conceptions of hepatitis is the presence of inflammatory cells in the portal connective tissue. With the exception of Mentzer [21] and Aiello [23] these studies have been made on surgical material in known cases of cholecystitis, and because in these cases this lesion was found it was reasoned that the lesion must be in some way associated with the etiology of cholecystitis, in spite of the fact that it is seen almost constantly by pathologists who examine routine sections of autopsy material. Mentzer[21] came to the conclusion that "60 to 70 per cent of all livers show evidence of pathological changes at postmortem examination, regardless of the presence or absence of gall-bladder disease." He goes on to say, however, that in the case of minor gross lesions of the gall-bladder, such as diverticula and cholesterosis, the percentage of liver involvement is about 60 "as compared with definitely inflammatory diseases, such as gall stones," where 97 per cent of the livers showed hepatitis. In this series of cases Mentzer's conclusions were not borne out.

This study revealed no definite explanation for the almost constant finding of inflammatory cells in the liver. Both Graham [5]

and Judd [16] admitted finding hepatitis identical with that seen in conjunction with cholecystitis in cases showing no evidence of gall-bladder infection, and one is forced to favor Martin's [22] idea that the inflammation is secondary to the lodgement of bacteria and toxic substances carried to the liver by the portal blood. This conception is given some support by the work of Andrews and Hrdina [28, 29] who cultured *B. Welchii* from the livers of normal dogs and showed that after the ligation of the cystic and common bile ducts in dogs, a cholecystitis was produced that first involved the portion of the gall-bladder adjacent to the liver. Arnold [30] found that in dogs with a loop of small intestine experimentally isolated and stimulated by the presence of food in a medium with a reaction of pH 8 the lymph from the thoracic duct contained 500 to 1000 bacteria per cc. All of these facts seem to favor the intestine as the possible source and the portal blood as the conducting medium for the infection of the liver. The suggestion that the liver may obtain bacteria, under both normal and pathological conditions, is by no means new. Adami,[31, 32] in 1898 studied autopsy specimens of liver and was able to show numerous microörganisms in cases of cirrhosis. He thought these bacteria were *B. coli*. He was able also to demonstrate bacterial shadows in cases showing no liver pathology, and he was of the opinion that they represented bacteria in the process of destruction by the endothelium of the liver.

As mentioned above, none of the cases under consideration here were cases of clinical cholecystitis and until such a series is included in this group no definite conclusions can be drawn concerning the degree of liver inflammation in clinical cholecystitis. Most workers agree, however, that even in the presence of severe gall-bladder infection, the liver lesions were sometimes minimal, and certainly the Grades 3 and 4 cholecystitis seen in this series are comparable in degree to many well established clinical cases of chronic cholecystitis. There seems to be, however, a definite difference between the cholecystitis found at autopsy and the clinical disease. This is perhaps best shown in the matter of age and sex distribution. The autopsy lesion certainly occurs more frequently in the younger age groups than does clinical cholecystitis and, whereas in postmortem material the disease occurs almost equally in the two sexes, clinically the female is affected at least twice as often as the male. In the case of the lesser degrees of infection in the autopsy cases some

doubt may be thrown on their interpretation as true or at least significant infections, although in most instances they are as marked as those seen in certain specimens removed surgically.

The occurrence of increased connective tissue in the portal spaces, as demonstrated in this series, was too infrequent to be of any practical importance in the consideration of the etiology of cirrhosis. In 17 cases such an increase was noted and in 5 of these cases the lesion was well advanced and presented the picture of typical portal cirrhosis. In the other 12 cases the increase in connective tissue was recognized only on microscopic examination.

Conclusions

1. Infiltration of the portal spaces by lymphocytes and polymorphonuclear leukocytes is seen almost constantly in the liver at autopsy.

2. This portal infiltration seems to have no particular relation to generalized infections or to any specific type of disease.

3. The portal infiltration apparently bears no relation to the non-clinical cholecystitis frequently found in routine postmortem examinations.

4. The cholecystitis found in routine postmortem examinations is seen less frequently in the female than clinical cholecystitis and it is more frequent in the younger age groups than clinical cholecystitis.

5. In this autopsy series cirrhosis occurs too rarely to justify the conclusion that the hepatitis described has any relation to the etiology of cirrhosis.

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DESCRIPTION OF PLATES

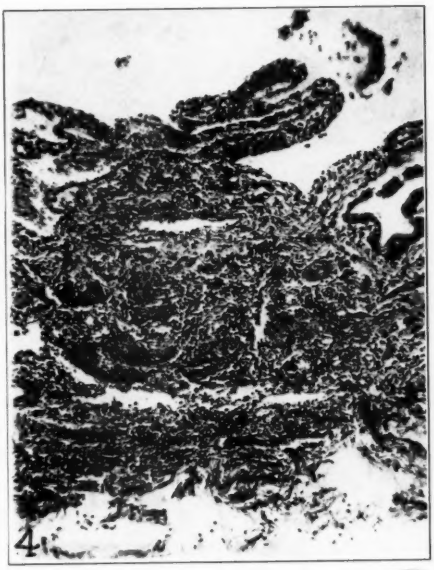
PLATE 70

Sections of the wall of the gall-bladder showing the respective degrees of cholecystitis. Figures 1, 2, 3, and 4 show respectively the corresponding grades of the inflammation.

1

2

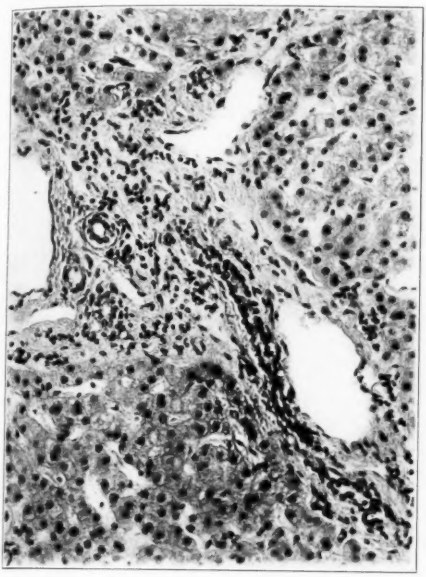
3



4

PLATE 71

Sections of the liver showing the degrees of portal infiltration. Figures 5, 6, 7 and 8 correspond respectively to Grades 1, 2, 3, and 4 of portal infiltration.



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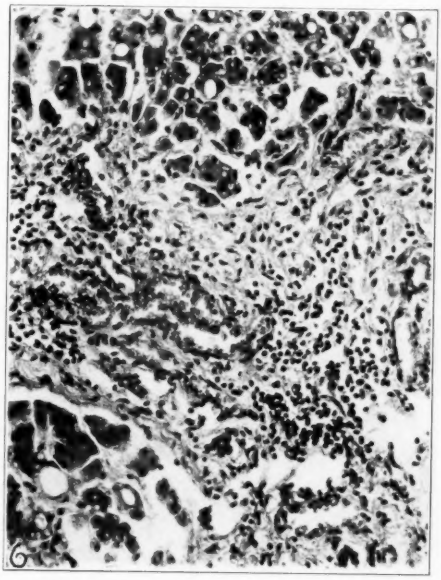

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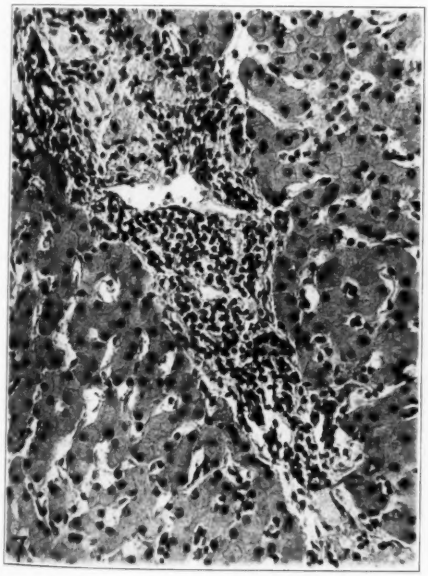

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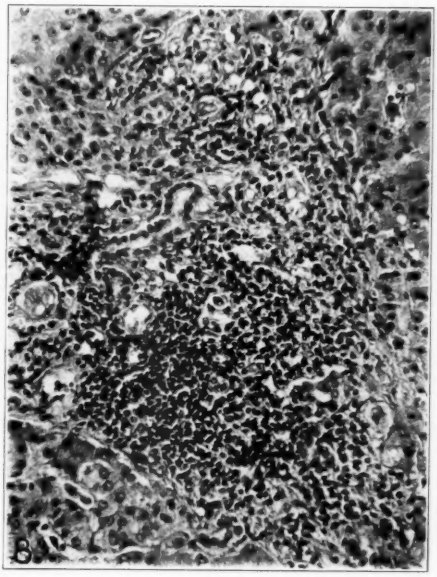

8

CALCIFIED EPITHELIOMA OF THE SKIN*

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INTRODUCTION

The so-called "calcified or calcifying epithelioma" of the skin is rarely mentioned in the English literature. I have been able to find only 1 case diagnosed as calcified epithelioma of the skin, reported by Nicholson in 1917, and 2 similar cases described as calcified adenoma of the skin, 1 reported by Eve and the other by Hutchinson. Even so comprehensive a work as Ewing's Neoplastic Diseases makes no mention of them. On the other hand, these tumors are not infrequently reported and discussed by German and French authors. In the Department of Pathology of Peiping Union Medical College 10 cases have come to our notice in the past 15 years, during which time more than 22,000 surgical specimens have been examined. In comparison with the ordinary malignant squamous cell epitheliomas and the epidermoid (sebaceous) cysts of the skin the infrequency of this type of tumor is remarkable and sufficient to warrant a report of this kind, especially in view of the scantiness of the English literature on the subject.

The detailed descriptions of our 10 cases are given in the case reports to follow. The important features that are presented in Table I can be summarized as follows: The cases are equally divided between two sexes, with an age incidence ranging from 13 to 29 years. One-half of the cases occur before the age of 20 years. In 7 of the cases the tumors are found on the head (5 on face, 2 on neck) and in 3 cases on the arm. The tumors are hard or firm, 0.5 to 9.5 cm. in diameter, sharply encapsulated, subcutaneously situated, freely movable over the deeper tissues but usually adherent to the overlying intact skin. Their cut surfaces show a characteristically sandy or gritty appearance.

Microscopically the parenchyma consists chiefly of degenerated epithelial cells, but isolated areas of living cells are usually to be found. These latter cells are small, oval, deeply staining and closely

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packed without distinct cell borders, but here and there they are squamous in shape with cornification and pearl formation. Mitotic figures are as a rule difficult to find. The degenerated cells stain a deep pink color in hematoxylin eosin sections and their outlines are generally well preserved. The stroma is composed of strands or a network of hyalinized or cellular fibrous tissue that varies in amount in different places and that is continuous with the fibrous capsule. Cholesterin crystals and other fatty substances may be found in the degenerated epithelium and in the stroma. Calcification is constantly present in the degenerated epithelium, either as fine dusting or as irregular lumps, and in the stroma either as incrustation of the fibrous fibers or as irregular granules. Bone tissue is frequently laid down from the periphery inward to replace the calcified epithelial masses. Surrounding the cholesterin crystals and calcified areas foreign body giant cells are frequently seen.

Review of Literature

Nomenclature: The names by which these tumors are designated by different authors who have reported them are not uniform. "Calcified epithelioma" ("verkalktes Epitheliom," "epithéliome calcifié") is by far the most popular name employed. They have been variously regarded as endotheliomas (Perthes); sebaceous gland adenomas (Eve, Barlow, Hutchinson); atheromas or epidermoids (Joannevics, Sternberg, Krüger (quoted from Gans), Virchow); carcinomas (Strassberg, Frey, Förster, von Noorden, Denecke, Bilke (quoted from Gans)); specially modified dermoids (Souligoux and Pilliét); or even as papillomas in dermoid cysts (Linser). That these authors are speaking about the same sort of tumor can be determined by reading their descriptions and examining their plates or illustrations. Perthes thinks that his "calcified endothelioma" is not different histologically from the "calcified epithelioma" of Malherbe and Chenantais, who published the first report in 1880, and attributes the discrepancy in name merely to difference in interpretation. Reverdin mentions the fact that 2 of his cases look like endotheliomas. Firket thinks that calcified epithelioma and endothelioma belong to the same category. Barlow thinks the "cancroids" of Förster, the "epitheliomas" of Chenantais and von Noorden, and the "sebaceous adenomas" of Eve and Hutchinson are the same thing and

TABLE I

Case No.	Sex	Age	Location	Duration	Diameter	Consistence	Free-movability	Adherence to overlying skin	Fibrous capsule	Epithelial parenchyma						Stroma			Fat				Cholesterin crystals		Calcification		Ossification	Cystic spaces
										Small, deeply-staining, without distinct cell borders	Squamous, with distinct cell borders	Mitoses	Epithelial pearls	Cornification	Necrotic cells	Hyalinized or cellular	Foreign body giant cells	Lymphocytic infiltration	In necrotic epithelium		In stroma		In necrotic parenchyma	In stroma	In parenchyma	In stroma		
																			Neutral	Fatty acid	Neutral	Fatty acid						
		yrs.		*yrs.*	*cm.*																							
1	M	26	Eyelid	1	1.5	Hard	?	?	+	+	−	−	+	+	+	H	+	+	o	o	o	o	−	−	+	+	−	+
2	M	29	Left temporal	1½	1.2	Hard	+	+	+	+	−	−	+	+	+	H	+	+	o	o	o	o	+	+	+	+	−	+
3	M	26	Right temporal	1	3.0	Hard	+	+	?	+	−	−	+	+	+	C	−	−	o	o	o	o	−	−	+	+	−	+
4	M	20	Neck and face	?	3.0	Hard	+	+	+	−	−	−	−	−	+	H	+	+	−	+	+	+	+	+	+	+	+	+
5	M	22	Right arm	2	9.5	Hard	+	+	+	+	+	−	+	+	+	H	+	−	−	+	−	+	−	−	+	+	+	+
6	F	13	Forehead	3	2.2	Firm	+	+	+	+	+	−	−	+	+	HC	+	−	−	+	−	+	−	−	+	+	−	+
7	F	23	Left arm	½	3.0	Firm	+	+	+	+	+	+	+	+	+	HC	+	+	−	+	−	+	−	+	+	+	−	−
8	F	18	Right temporal	4	2.5	Hard	+	−	+	−	−	..	+	+	+	H	+	+	o	o	o	o	−	−	+	+	+	−
9	F	16	Left arm	¼	0.5	Firm	+	+	+	+	+	−	+	+	+	C	+	−	o	o	o	o	−	−	+	−	−	−
10	F	20	Left mastoid region	8–9	1.2	Firm	+	−	+	−	−	..	−	−	+	C	+	+	o	o	o	o	+	+	+	+	+	−

o = No tissue available for fat reaction. H = Hyalinized. C = Cellular.

prefers the last-mentioned name. Malherbe and Chenantais call these tumors "calcified epitheliomas of the sebaceous gland," and Saltykow uses the name "calcified epithelioma" for a type of tumor that he thinks arises from sebaceous gland anlage. Frey designates his tumor "psammocarcinoma of the skin." Gans distinguishes such tumors from true squamous cell epitheliomas with calcification or ossification by the presence of a capsule in the former and its absence in the latter. He also thinks that a so-called calcified epithelioma, before it begins to calcify, is histologically identical with a basal cell carcinoma, but this diagnosis is excluded by the sharp encapsulation of the tumor. Saltykow also shares this view. Strassberg and Dubreuilh and Cazenave are of the opinion that "calcified epithelioma" is not a good name because the calcification, being observed also in the common squamous epitheliomas of the skin, is not a characteristic process; it is the necrosis alone that is the essential feature. Bilke also thinks of calcification as a secondary process. Since the name "calcified epithelioma" has already been universally accepted to denote a clinically well defined type of tumors, it may be conserved (Dubreuilh and Cazenave).

Histogenesis: These tumors are by the consensus of opinion traced to an epithelial genesis. They are believed by different authors to arise from preformed glands, dystopic deep-lying epithelial cells and misplaced anlage material (Kaufmann). Saltykow, Firket, Murakami and Lücke favor the origin from epithelial cell rests. Reverdin thinks that the ectodermal inclusion may be of either embryonal or traumatic nature. Jadassohn (referred to by Gans), Dössekker, Bilke, and in certain respects also Firket, Murakami and Saltykow, believe in a sebaceous gland anlage as the source of these growths. Frey considers a kind of embryonic rest of primitive sebaceous gland and primitive squamous epithelium, with the double potentiality of forming fat cells and cornified epithelium in an imperfect and disturbed fashion. This forms a mixed product of incompletely differentiated fat and epithelial cells, which in time are overtaken by calcification. Sternberg, Krüger, Joannevics and Virchow take these tumors to be of the same origin as the epidermoids (sebaceous cysts), but Gans and Bilke are opposed to this view because these tumors, besides the difference in anatomical structures, occur too rarely, as compared with the exceeding frequency of the epidermoids (sebaceous cysts). Jadassohn thinks they are closely related to nevi.

TABLE II

Author	No. of cases	Year
Perthes	27	1894
von Noorden	19	1887
Malherbe	18	1881
Dössekker	11	1921
Saltykow	5	1913
Murakami	4	1911
Henzi	4	1914
Bilke	4	1922
Stieda	3	1896
Pilliét	2	1890
Thorn	2	1898
Strassberg	2	1911
Lücke	1	1863
Eve	1	1882
Hutchinson	1	1890
Souligoux and Pilliét	1	1898
Reverdin	1	1901
Cornil	1	1904
Sternberg	1	1904
Caubet	1	1905
Lapointe	1	1907
Sehrt	1	1910
Firket	1	1912
Nicholson	1	1917
Frey	1	1920
Léjard (Henzi)	1	Not given
Sokolosky (Henzi)	1	Not given
Total	116	

Bilke believes they have a marked similarity to cholesteatomas that arise from epidermal inclusions in the central nervous system, and his belief was strengthened when he found clearly lamellated cornified masses in such tumors (Gromiko). Souligoux and Pilliét regard them as related to dermoid cysts because both occur more frequently in young people, and Linser goes so far as to consider a calcified epithelioma as nothing more than a papilloma growing in a closed cavity of a dermoid cyst and filling up the entire cavity and adhering to its walls. Gans thinks that those tumors whose glandular origin is evident are best considered as adenomas or nevi, while a classification for others whose origin is doubtful should be postponed until the studies of the earlier stages of the tumor can be made.

Analysis of Reported Cases as to Body Regions, Sex, and Age: From the literature available to me, 116 reported cases have been collected and are presented in the following table (Table II).

TABLE III

Head and neck	Eyebrow, eyelid or orbit	12 cases
	Neck	9 "
	Forehead	4 "
	Cheek	3 "
	Lower jaw	2 "
	Face	2 "
	Parotid region	2 "
	Ear lobe	2 "
	Temporal region	1 case
	Occipital region	1 "
	Mastoid region	1 "
	Lower lip	1 "
	Scalp	1 "
	Exact location not specified	16 cases
	Total	57 cases

Table III (*Continued*)

Upper extremity	Arm	11 cases
	Forearm	6 "
	Cubital region	1 case
	Elbow	1 "
	Total	19 cases
Trunk	Back	8 cases
	Breast or nipple region	3 "
	Pectoral region	1 case
	Angle of scapula	1 "
	Lower back	1 "
	Lumbar region	1 "
	Total	15 cases
Lower extremity	Thigh	2 cases
	Leg	1 case
	Foot	1 "
	Total	4 cases
	Regions not mentioned	21 cases
	Total	116 cases

The tumors reported in literature are most frequently located on the head and neck, as shown in Table III. Although such tumors usually occur singly, two or more similar growths are occasionally found in different regions of the same individual (Perthes).

Of the 116 reported cases only 35 are found in which the sex of the patient is mentioned. Of these, 23 cases occurred in the female and 12 in the male. Malherbe has the impression that the female sex is predisposed.

There are 31 cases in which the age of the patient is known and they are distributed as follows: 14 below 20 years of age, 8 between 21 and 50, and 9 after 51. Of the 14 cases occurring before 20, 7 are

before 10, and 3 of these 7 before 16 months. The youngest case, reported by Firket, was an infant 2 months old. The view that these tumors are most frequently encountered in youth is shared by Malherbe, Firket, Souligoux and Pilliét, and Reverdin. On the other hand, Kaufmann and Perthes are of the opinion that they occur at any age without special predilection.

Predisposing Causes: The direct cause of these tumors is unknown. Malherbe has three times observed trauma as the exciting cause. In one case the tumor arose after the sting of a wasp.

Description of the Tumor: The calcified epitheliomas constitute a special tumor group by their structure and location. Their appearance is so unique that, if one has not known of such a tumor as calcified epithelioma, the diagnosis is difficult (Saltykow). They form the immense majority of the bony tumors of the skin (Malherbe). They are reported as lying in the cutis, subcutis, or partly in the cutis and partly in the subcutis. They are slow-growing, rounded, stone-hard, cartilaginous or soft, freely movable, sharply encapsulated, at times cystic, and usually have a gritty cut surface. They are usually small, described as hazel-nut or walnut-sized growths. The smallest reported is the case of Sehrt, where the tumor measured only 4 mm. in diameter, while both Murakami and Gromiko have described tumors as large as the head of a newborn. The tumor consists of a connective tissue capsule and stroma with epithelial cells that are in the form of nests, lobulated masses, or in alveolar arrangement.

(*a*) *The Epithelial Cells:* The epithelial cells are described as typical squamous cells with keratohyalinization and pearl formation (Saltykow, Firket, Kaufmann, Sehrt, Gromiko). According to Eve, however, they are small, round, with deeply staining nuclei, closely packed together with no intercellular substance. Gans finds that before calcification they appear like the cells of a basal cell carcinoma, and he distinguishes in the cell masses an outer and an inner layer of cells, those of the outer layer being more cubical and those of the inner layer more rounded.

The characteristic tendency of the epithelial cells to undergo necrosis has been described by many authors. The necrosis begins usually in the centers of the masses and spreads toward the periphery. The original outline and structural details of the necrotic cells are generally well preserved. In the necrotic epithelial masses

one finds cholesterin crystals, fatty acids and calcium deposits. The latter are either in the form of deeply stained masses or concentric layers, or merely as a fine dusting of the cell cytoplasm.

Cornification of epithelium is, according to Gromiko, a typical feature of this tumor. The cornified epithelium of the calcified epithelioma is distinguished by Thorn from the epithelial pearls of the squamous cell epithelioma by the lack of any definite structure in the former and the presence of nucleated layers of corneum in the latter. Murakami could not find this difference, while Gromiko observed some cornified pearls of the calcified epithelioma quite similar to, but others (being much larger and irregular) different from, those of the squamous cell epithelioma.

(*b*) *The Connective Tissue Stroma:* The connective tissue stroma is described typically as fibrillar or acellular. It extends from the capsule into the interior of the tumor in the form of septa, strands, or a sort of network, in the meshes of which lie the epithelial masses. The connective tissue fibers may also undergo necrosis and those fibers adjacent to the calcified epithelial masses may also become incrusted with calcium. Cholesterin crystals are found in the degenerated stroma. The stroma frequently shows edematous and hyaline changes, and rarely mucinous and fibrosarcomatous degeneration (Léjard, quoted from Henzi). Foreign body giant cells are usually found around the calcified masses and cholesterin crystals.

(*c*) *Cystic Spaces in the Tumor:* Cystic spaces or cavities are generally found in the tumor as a result of degeneration or liquefaction of epithelial cells (Kaufmann); edema of the connective tissue (Murakami); shrinkage of volume consequent upon cornification, calcification, and disappearance of tissue fluid (Gans); or contraction following fixation (von Noorden, Gromiko).

(*d*) *The Peripheral Parts of the Tumor:* The peripheral parts or the immediate vicinity of the tumor, according to Gans and others, show no inflammatory reaction. Such a reaction, however, has been described by Henzi.

(*e*) *The Skin Overlying the Tumor:* Aside from flattening and stretching consequent upon the expansile growth of the tumor, the overlying skin is not ulcerated or invaded even when the tumor may reach a great size (Gromiko).

Necrosis and Calcification: The great tendency of these tumors to necrosis is ascribed by some to their poor blood supply (Nicholson,

Gromiko, Hutchinson, Sehrt), and by others to the low vitality of their cells (Gromiko). It is generally agreed that calcification follows necrosis. There is reason to believe that, in some cases, calcification is preceded by deposition in the necrotic tissue of fatty acids, which form insoluble soaps with the calcium of the blood or tissue fluid.

Ossification: Ossification is looked upon as a natural sequela of necrosis (Saltykow) and as imparting no additional distinctive feature to the tumor (Malherbe). Wilkens, 1858 and Denecke, 1893 are the earliest authors to describe bone formation in calcified epitheliomas, then called "carcinomas." The bone formation is described as perfectly well formed with lacunae, canaliculi, Haversian systems and marrow cavities. Many authors report bone occurring along the edges of the calcified epithelial masses and in the stroma. The bone formation is preceded by the formation of a granulation tissue that contains embryonic fibroblasts with osteoblastic function (Nicholson). Bone forms at the site of the calcified deposits, which are first dissolved and removed by the giant cells (Strassberg, Saltykow, Denecke, Walkoff, Henzi). Actual marrow elements, *e.g.* eosinophilic and neutrophilic myelocytes and leukocytes (Sehrt); megakaryocytes (Nicholson); nucleated red cells (M. B. Schmidt); and fat cells (Saltykow), have been found in the stroma and in the marrow cavities.

Clinical Behavior: Calcified epitheliomas are generally benign, non-destructive and non-metastasizing. Malherbe says this tumor is almost always benign and he did not know of more than 2 cases with recurrence after operative removal. Reverdin found only 2 recurrent cases out of 53 published observations. Gromiko reports a case with malignant degeneration in a workman 32 years of age. The first tumor, situated on the upper third of the right forearm, was a small, slow-growing, rounded mass that was removed 2 years after it had been noticed. Cut surface of this tumor showed small grains like sand, but no histological examination was made. Nine years after the operation a new tumor appeared on the inner side of the right elbow, growing steadily from a small nodule to the size of the head of a newborn. No history of trauma was given. The consistence of the tumor was hard, and the capsule adherent to deeper tissues. The clinical diagnosis was "sarcoma of right cubital region." The pathological diagnosis was "calcified epithelioma."

Ossification was present. Five months and 8 days after the second operation the patient again noticed a tumor, appearing at the site of the operative scar, that grew rapidly and caused much pain. Pain was also felt in the fourth and fifth fingers of the right hand. The tumor was not movable and amputation of the humerus was performed. The diagnosis was: "calcified epithelioma of skin with malignant degeneration and recurrence." This last tumor showed calcification and ossification in small amounts. Both the second and the third tumors were hemorrhagic, especially the latter. Inside the blood vessels of the stroma and the capsule of both tumors there were found tumor cells that also underwent massive cornification in the same manner as the cells of the tumor parenchyma. Mitotic figures were found in the epithelial cells of both tumors. Gromiko explains the recurrence by the probable presence of tumor cells inside blood vessels in the close neighborhood of the tumor, which were not removed with the tumor mass during operation. According to him, Thorn has also found the tumor cells inside the blood vessels and Denecke has found them inside the lymphatics in the stroma of the tumor.

Case Reports

Case 1: S-10,122. Specimen sent in from Good Will Hospital, Nanhsuchow, Anhwei, on Dec. 6, 1926. Chinese male, aged 26 years. Tumor of eyelid of 1 year's duration.

Pathological Report: Specimen consists of a piece of tissue measuring about 1.5 cm. in diameter. Consistence hard. On cut section the tissue is grayish white and whitish opaque in color. (No gross specimen has been left; this description is from the routine record.)

Microscopic Examination: There is a fibrous capsule from which trabeculae of hyalinized stroma ramify into the interior of the mass. In between the hyalinized stroma lie masses of epithelial cells that are totally necrotic with nuclei and cytoplasm staining alike with eosin, but whose structural details are remarkably well preserved. Pearl formation and cornification are frequent in these masses. In places the central parts of these dead epithelial masses do not take any stain and are in the form of a very pale granular débris. Here and there among the dead masses one finds, however, isolated islands of living epithelial cells whose nuclei are small, oval or spherical, dark blue-staining, and closely packed in the form of a syncy-

tium without visible cell borders. Transitions, either abrupt or gradual, from these living epithelial cells to the dead cells, are readily found. Numerous giant cells are found at the edges of the necrotic epithelial masses; some are also found free in the stroma tissue. Calcification is seen in the epithelial masses in the form of small dense masses or as irregular, scattered areas. Calcification of the stroma is in the form of incrustation of the connective tissue fibrils. The stroma is infiltrated with small lymphoid cells, especially near the periphery. Many irregular, empty or cystic spaces are found in the tumor.

CASE 2: O. P. D. No. 126348, S-12,408. C. S. J., a Chinese male clerk, aged 39 years, came to Peiping Union Medical College on April 3, 1928, with the complaint of a swelling in the left temporal scalp for 1½ years. It had begun without known cause 18 months before as a small, hard, slipping mass which grew bigger gradually. It had never been painful or tender. Upon examination the swelling was found to be a very hard, non-tender, flattened and freely movable tumor mass 1.5 by 1.5 cm. in diameter. It seemed to be attached to the overlying skin.

Pathological Report: Specimen consists of a nodular mass measuring 1.2 by 0.5 by 0.3 cm. Its external surface is grayish pink in color and in places shows light yellow, elevated nodules. Consistence hard. On section the cut surface is grayish pink with light yellow areas.

Microscopic Examination: The tumor is surrounded by a thin fibrous capsule and consists of a network of ramifying bands of fibrous tissue stroma, in the meshes of which lie necrotic epithelial masses. Although totally necrotic, as judged from the staining qualities, these cells preserve their outline and structural details strikingly well. The cells are small and of squamous type with rather extensive cornification and formation of pearls. Only one of these necrotic masses has a narrow strip of living cells attached to its edge. These living cells are small, oval, closely packed, with deeply staining nuclei without distinct cell borders, and are sharply demarcated from the stroma. These cells resemble the basal cells of the epidermis. Calcification of the necrotic epithelial masses and cornified pearls is extensive and occurs as deep blue areas with stratification, as irregular masses or coarse granules, or as very fine dust particles in the cytoplasm of the dead epithelial cells, whose outlines are still preserved. Calcium granules are also found in the connective tissue fibers that may become themselves so heavily incrusted

with calcium that they appear as coarse, blue, wavy lines. Cholesterin crystals are occasionally found in the necrotic cell masses and frequently found in the degenerated stroma which also shows edema and hyalinization. Giant cells are found at the edges of the necrotic and calcified epithelial masses and also scattered in the stroma. The stroma of the peripheral portion of the tumor is more vascular and more cellular, and infiltrated with more lymphocytes and wandering cells than that of the central portion. Cystic spaces are present apparently as a result of necrosis and degeneration.

CASE 3: O. P. D. No. 129519, S–12,641. P. Y. M., a Chinese policeman, aged 26 years, came to Peiping Union Medical College on May 17, 1928, with the complaint of a painless swelling in the right temporal region for 1 year. Upon examination a tumor of the size of a small walnut was found; it was adherent to the skin but free from the bone of the skull. It was hard or cartilaginous, but soft in places. The diagnosis of sebaceous cyst was made and the tumor excised.

Pathological Report: Specimen consists of several irregular pieces of tissue measuring 0.2 to 1 cm. in diameter. Their external surfaces are rough, and are grayish pink and light yellow in color. The consistence is firm. On section the cut surfaces are grayish white in color and show light yellow, chalky, calcified areas.

Microscopic Examination: The growth consists of ramifying strands of fibrous tissue in the meshes of which lie necrotic masses of epithelial cells of squamous type with rather extensive cornification and pearl formation in their central parts. Although the epithelial cells are necrotic and their nuclei fail to stain, their outline and structural details are still preserved. Only in two or three places are small masses of living epithelial cells found. These are small, oval, with deeply staining nuclei, closely packed and without clear cell borders. They resemble closely the cells of the basal layer of the epidermis. Calcium deposits, often in the form of large and irregular masses, are found in the necrotic epithelial masses. All the cornified pearls are necrotic and without nuclei. Many of them are hyalinized, others are either completely or partially calcified. Irregular masses and coarse granules of calcium are found in the otherwise unchanged stroma and there is also incrustation of the connective tissue fibers with calcium. No giant cells are found. Cellular infiltration is absent in the peripheral as well as in the central region. Cystic spaces are occasionally found.

CASE 4: O. P. D. No. 134730, S-13,013. H. F. Y., a Chinese shoemaker, aged 20 years, came in on July 27, 1928 with the complaint of a rectal abscess. He was accidentally found to have unusual tumor nodules in the posterior cervical region and anterior to the left ear. No history was obtainable about these nodules. They were flattened, very hard or cartilaginous in consistence, freely movable but attached to the overlying skin. One such nodule was excised for biopsy.

Pathological Report: Specimen consists of a hard nodule measuring 3 by 1.5 by 1 cm., well encapsulated and covered in places with subcutaneous fat. On section the cut surface is grayish white in color, showing ramifying strands of bone-like tissue.

Microscopic Examination: The microscopic picture is in many respects identical with that of Case 5 (see below) which was better preserved and therefore will be more fully described. The tumor is encapsulated with fibrous tissue and consists of ramifying strands of hyalinized, fibrillar connective tissue stroma, in the meshes of which lie epithelial cell masses. There are no living, unaltered epithelial cells left. All the epithelial masses are totally necrotic, but their outlines and structural details are still preserved. Neither cornification nor pearl formation is seen. Many of the cell masses are partly or wholly calcified (see below), and the connective tissue fibers of the stroma are either incrusted with calcium or sprinkled over with granules of calcium. Cholesterin crystals are found both in the necrotic epithelial masses and especially in the degenerated stroma. The amount of fatty substance in the tumor is worthy of remark. By the scharlach R stain all the necrotic epithelial cells are, without exception, loaded with fine droplets of fat in their cytoplasm, while the entire stroma shows diffuse infiltration with large globules of fat. By differential stains (Nile blue and neutral red), the fatty acids are found to constitute the whole of the fatty material found in the epithelial masses and a large part of that found in the stroma. Neutral fat is present only in small amounts in the stroma and entirely absent in the epithelial masses. Giant cells are found in the stroma, but especially at the edges of the necrotic and calcified cell nests. Bone spicules with marrow cavities filled only with fatty tissue are found at the periphery, and sometimes entirely replacing the calcified masses. Cystic spaces are found in the tumor. The stroma at the periphery of the tumor and in the region of ossification is more cellular and vascular than elsewhere. Infiltration of lymphocytes and wandering cells is more marked at the periphery than in the central part of the growth.

CASE 5: O. P. D. No. 200170, S–19,525. C. W. K., a Chinese handicraftsman, aged 22 years, came in on June 9, 1931 with the complaint of a tumor of the right arm that had started spontaneously 2 years ago as a painless, small firm mass. It had grown in size rather quickly and was now as large as a fist. Upon examination the tumor was found to be nodular, hard, and slightly tender on pressure. Although freely movable over the muscle and bone it was adherent to the overlying skin in places. A diagnosis of fibroma was made and the tumor was excised.

Pathological Report: Specimen consists of a tumor mass having the shape and approximate size of a goose-egg, measuring 9.5 by 7.5 by 6 cm. It is heavy, stony in consistence, and sharply circumscribed, being covered on one surface by skin and on the other by subcutaneous fibrous tissue. The skin in some places can be slipped over the surface of the tumor but in other places it is adherent. Over the center of the adherent area there is in the skin a small round ulcer 3 mm. in diameter, that extends into a cavity in the substance of the tumor, from which it discharges a dirty, yellowish brown fluid. Except for the presence of this ulcer the epidermis of the skin overlying the tumor is entirely normal. The mass cuts like bone and cannot be sectioned except by sawing. The cut surface shows that the tumor, though largely in the subcutaneous tissue, also involves at certain areas the cutis — hence adhesion to the skin. The tumor tissue is grayish and yellowish white in color, sandy or chalky in consistence. The skin ulcer communicates with a cavity in the tumor 2 cm. in diameter, containing some grayish yellow, semiliquid material.

Microscopic Examination: The growth is well encapsulated by fibrous tissue. From the capsule ramifying strands of connective tissue extend into the center of the tumor forming a network, in the meshes of which lie irregular columns, masses or strands of epithelial cells. Over a few areas living epithelial cells are seen. They are small, oval, deeply staining and closely packed with vesicular nuclei, each of which contains a dense, dark blue nucleolus. These cells form almost syncytial masses with no clear cell borders. Those in the central part of the mass are more spherical and those of the periphery more cylindrical or flattened. Thus to a certain extent this tumor resembles the common basal cell epithelioma. However, only rarely can one find living cell masses without regressive changes. Almost always the central parts, and very frequently the entire masses, are necrotic, but the epithelial cells, in spite of the necrosis, retain their outline and structural details very well. Not infre-

quently one finds differentiation of the basal-like cells into pale, large, typical squamous cells, and further into cornified epithelial cells, some of which form regular pearls. The cornified pearls are practically all necrotic and are different from the epithelial pearls of the ordinary squamous cell epithelioma in that the latter consist of lamellated keratin with distinct, blue-staining nuclei, while the former have lost all nuclei and layers. Most of the necrotic masses undergo more or less complete calcification. Sometimes only one part of a necrotic cell mass is calcified and the boundary between the calcified and the uncalcified part is very sharp. Under high power distinct, fine calcium granules are seen in the cytoplasm of the necrotic cells, whose outlines are still preserved. Calcification also occurs in the stroma. The connective tissue fibers adjacent to the calcified epithelial masses become incrusted with calcium and appear as coarse, blue, wavy lines, or the hyalinized stroma between the calcified epithelial masses becomes sprinkled with deep blue, more or less uniformly-sized granules of calcium. The stroma consists of an acellular fibrillar connective tissue that is mostly hyalinized and also in places shows fairly marked edema, its fibrils being split apart by coagulated fluid that appears as fine, pink, granular material in the meshes. Cholesterin crystals are not found in the epithelial masses or in the stroma, but there is a large amount of fatty acid both in it and the degenerated epithelial cells. Neutral fat is almost entirely absent, even in the stroma, as shown by Nile blue and neutral red stains. Giant cells of foreign body type are frequent in the connective tissue, but especially numerous at the periphery of the necrotic epithelial masses, both calcified and uncalcified. Cystic spaces are present in the stroma, between it and the epithelial masses, or inside the necrotic epithelial masses, evidently as a result of necrosis and liquefaction of epithelial masses and edema of the connective tissue. The stroma of the peripheral parts of the tumor, as well as the connective tissue around it — for example in the cutis — is more cellular, more vascular, and infiltrated with more lymphocytes and wandering cells than it is in the central part of the growth, which is avascular, acellular and hyalinized. Where there is ossification the stroma has a marked cellular appearance and contains a great number of capillaries. Spicules of bone with well differentiated bone corpuscles and bone matrix are closely applied to the surface of the calcified masses. The bone forms a thin fringe

around a calcified cell mass, or replaces the outer half of the mass with a central inclusion of calcified epithelial cells whose outlines are still visible, or completely replaces the entire mass, in which case there appear also marrow cavities containing blood capillaries and loose fibrous tissue, but no myeloid or erythroblastic elements. All these various stages of ossification demonstrate clearly that bony tissue is not formed in the stroma but in the calcified epithelial masses, from outside inward. Giant cells are numerous at the edges of the calcified cell masses, but are never found around the bone.

Section of the ulcer of skin and the edge of the cavity shows just an ordinary abscess cavity lined by granulation tissue densely infiltrated with polymorphonuclear leukocytes.

CASE 6: S–20,857. Specimen received in January, 1932, from Weihwei Hospital, Honan. C. C. C., a Chinese female, aged 13 years, had had a small nodule on the forehead for 3 years, growing rapidly during the last half year. On examination a well defined, firm nodule was found partly in the skin and partly in the subcutaneous tissue. A diagnosis of sebaceous cyst was made and the tumor was removed.

Pathological Report: Specimen consists of a well encapsulated nodule, measuring 2.2 by 2.2 by 1.5 cm., situated beneath the intact skin. Consistence fairly firm. The cut surface shows whitish, chalky, very firm strands and masses separated by soft, edematous, grayish tissue.

Microscopic Examination: The growth has a well defined fibrous capsule and consists of irregular masses and strands of epithelial cells, among which no mitotic figures have been found. The epithelial cells consist in some places, of small, oval, deeply staining nuclei, each with a nucleolus. These nuclei are closely packed together so that cell borders are not seen, but in most places the nuclei are larger and the cell borders are sharply defined, in contrast with the very faintly stained or transparent cytoplasm. In some areas these latter cells are swollen with a distinctly vacuolar cytoplasm; sometimes the vacuoles even push the nucleus into an eccentric position. However, the scharlach R stain reveals that these cells contain no more fat than the unchanged small cells, although much fat in the form of big droplets is present in the stroma and in the degenerated epithelial cells. Most of the cell masses are totally necrotic, others are partially so in the center. The structural details of the necrotic cells are remarkably well preserved. Calcium is present in the ne-

crotic masses as heavy, sharply demarcated, irregular masses staining deep blue with hematoxylin, or as fine granules diffusely scattered in the cytoplasm of the individual necrotic cells, appearing as bluish dust in the low power field. The stroma consists of fibrous tissue that is more cellular in the periphery than in the center of the tumor and is in places hyalinized. Foreign body giant cells are present in the stroma, but especially along the margin of the necrotic epithelial or calcified masses. Calcification in the stroma is found either as incrustation of the fibers of the fibrillar stroma tissue, which appear as heavy, dark blue wavy lines, or as solid irregular masses or small granules deposited here and there in the stroma. Cystic spaces are found in the stroma and between the stroma and the epithelial masses. No cholesterin crystals are found in the stroma or in the degenerated epithelial masses.

CASE 7: O. P. D. No. 240400, S–21,270. W. C. H., a Chinese housewife, aged 23 years, came in on April 2, 1932, with the complaint of a slowly growing, soft mass on the left arm for about 5 months. The mass started as a small, firm, painless nodule under the skin. It ruptured spontaneously and discharged some pinkish fluid. The rupture closed up spontaneously in a week's time and the mass grew slowly but steadily, more rapidly in the last 2 months, though still painless. No similar growth elsewhere on the body. Upon examination a freely movable, globular, subcutaneous mass, soft and non-tender, 3 by 2.5 cm., was found over the anterior aspect of the left arm. The overlying skin was thin, pinkish in color and adherent, but not ulcerated. A diagnosis of parasitic cyst was made and the growth was excised.

Pathological Report: Specimen consists of a nodular mass, measuring 4.5 by 3 by 2.7 cm., covered externally by skin that is loose, wrinkled and freely movable. The inner surface of the tumor is covered by subcutaneous fat tissue. Consistence firm. On section a sharply circumscribed and encapsulated firm nodule, measuring 3 by 2 by 2 cm., is found in the subcutaneous tissue. Between it and the overlying skin there is a layer of loose fibrous tissue. The cut surface of the tumor shows whitish gray and chalky, opaque, ramifying strands and masses with some dark red streaks near the periphery.

Microscopic Examination: The tumor is completely invested by a well defined fibrous capsule and consists of irregular masses or islands of epithelial cells, between which there is a small amount of fibrous stroma moderately infiltrated with lymphocytes and in places hyalinized. The majority of the epithelial cell masses are necrotic, the cell nuclei staining pale pink like the cytoplasm. Corni-

fied pearls are frequently found in these masses. The structural outlines of the epithelial cells are well preserved in spite of necrosis. Occasionally an entire mass is seen to have undergone complete cornification, appearing as a heap of granular or structureless material poorly stained. Numerous giant cells are at the edges of the necrotic epithelial masses. Living epithelial masses are also found, especially near the capsule. They are composed of small, oval or spherical undifferentiated cells, with deeply staining nuclei closely packed without visible cell borders in the form of a syncytium. Necrosis is frequent in these masses, and usually one finds a pink necrotic central region surrounded with a blue-staining living peripheral zone. An exceptional feature of this tumor is the presence of a large number of mitotic figures in the living epithelial cells which are not found in any other tumor of the series reported. The transition from the small, dark blue, undifferentiated living cells to the pink-staining, necrotic squamous cells is either gradual or rather abrupt. Occasionally leukocytic infiltration is present in the necrotic part of the tumor. It is of interest to note that calcification of the necrotic masses occurs only to a slight extent. Most of them remain free. Calcification occurs in the form of scattered, irregular, heavy lumps in the dead epithelium, or less often as fine granulation or dusting of the cytoplasm of the epithelial cells. In the stroma calcification in the form of irregular masses or incrustation of fibrils is rare. Giant cells are present in great numbers and cholesterin crystals are not infrequently found. The Sudan III stain shows a large quantity of fat droplets in the degenerated epithelium, but not in the living cells. Large, scattered fat droplets are also seen in the stroma.

CASE 8*: S–21,311. D. T. W., a Chinese female, aged 18 years, had a stony-hard, freely movable subcutaneous tumor in the right temporal region. Four years previously she had had a boil over that region which, after a while, healed spontaneously with the formation of a very small, flat, smooth scar. In this scar, however, she later discovered a small nodule, firm and freely movable, that grew in size very slowly but continuously until at the time of excision it was 2.5 by 1.7 by 0.8 cm.

Microscopic Examination: The tumor mass consists of irregular masses of epithelial cells embedded in a fibrous stroma that is in a few places cellular but in most places dense, collagenous or hyalin-

* Only a paraffin section slide of the tumor growth was received on April 7, 1932, from Dr. H. C. Pai of the Mukden Medical College, with the above brief clinical note.

ized. The block represents only a small portion of the tumor and only at one side is the tumor tissue surrounded by a thin layer of fibrous capsule and some fibro-adipose tissue. Beneath this ill defined capsule there is dense lymphocytic infiltration of the tumor stroma. The epithelial masses are all totally necrotic with pink-staining nuclei, and all show calcification of varying degrees, except one or two. No living epithelial cells are present. In the necrotic epithelial cell masses cornified pearls are frequently found, thus indicating their squamous nature. The outlines of the cells are still recognizable, although they are necrotic. Calcification occurs from mere dusting or fine granulation of the cells to a massive, deep blue deposit obscuring entirely the cellular structure of the masses. Giant cells of foreign body type are seen at the periphery of the calcified masses, many of which are completely or partly surrounded by a thin fringe or a thick layer of typical bone. Sometimes the calcified mass is almost completely replaced by bone. The structure of the epithelial cells of the calcified masses is well preserved, even when their outer zone has been changed into well formed bone. Calcification of the stroma occurs chiefly in the form of incrustation of the hyalinized fibrils. Giant cells are occasionally found in the stroma.

CASE 9: S–21,946. W. M. Y., a Chinese female, aged 16 years. A firm, freely movable, subcutaneous mass, 0.5 cm. in diameter, over posterior lateral aspect of left arm, somewhat adherent to the overlying skin, of more than 3 months duration. Tumor excised.

Pathological Report: Specimen consists of 3 broken pieces of tumor tissue measuring 0.5 cm. each in length. Two of them externally are partly covered by epithelium and partly by fibrous tissue. The third piece is covered by adipose and fibrous tissue. Consistence firm. Cut surfaces show yellowish, opaque chalky strands embedded in a whitish tissue mottled with a few small dark red areas and encapsulated on the free edge of epithelium or fibro-adipose tissue.

Microscopic Examination: The tumor tissue consists of irregular strands or masses of totally necrotic, pink-staining squamous epithelium with areas of cornification and pearl formation. The structures of the squamous cells are preserved in spite of necrosis. Small, living epithelial cell masses, few in number, are found here and there, consisting of dark blue-staining, small, oval nuclei, each with a single nucleolus, closely packed in purplish-staining cytoplasm in the form of a syncytium without clear cell borders. The central

parts of these masses practically always show necrosis. Oftentimes necrotic masses have only a fringe of living cells at the periphery or to one side. The transition between the living and necrotic cells is gradual, much more gradual than in the previous cases. The dark, small, closely packed nuclei differentiate into larger, paler and less closely grouped oval or rounded nuclei, each with one and sometimes two nucleoli, and as they differentiate the cytoplasm increases in amount and the cell borders become definite. Masses of these relatively well differentiated, living squamous cells are present between the small typical living cells and the necrotic squamous epithelium, and constitute a rather special feature of this tumor because, in the previous cases, these cells were very few or not seen. Toward the necrotic part of the tumor the nuclei of the cells become pyknotic until in the necrotic area they stain a pale pinkish color of the same degree as the cytoplasm. The fibrous stroma is rather cellular and contains foreign body giant cells. Numerous areas of calcification are seen in the necrotic cell masses. The calcium is seen under higher magnification as irregular granules or fine dusting in the cytoplasm of the cells. There is no bone formation.

CASE 10: O.P.D. No. 246613, S–22,034. Y. S. F., Chinese female, a student, aged 20 years, came in on July 1, 1932, with the complaint of a tumor in the left side of the neck of 8 to 9 years duration, and of slow growth. Examination showed a freely movable, subcutaneous, firm, slightly tender tumor nodule the size of a pea, situated behind the left ear lobule in the mastoid region. A diagnosis of calcified sebaceous cyst was made and the growth excised.

Pathological Report: Specimen consists of an irregular nodule of bony-hard consistence, measuring 1.2 by 0.8 by 0.4 cm. The external surface is smooth and is covered by congested, purplish, fibrous tissue. The mass could not be sectioned before decalcification. After that the cut surface revealed interlacing or anastomosing strands of yellowish brown and grayish, opaque chalky tissue.

Microscopic Examination: The tumor is enveloped by a well defined, thin, fibrous capsule, and consists of irregular anastomosing strands and masses of necrotic and mostly calcified squamous epithelial cells, the structural details of which are still fully recognizable in spite of necrosis and calcification. No epithelial pearls are found and no living epithelial cell masses are present. Calcification of the necrotic masses is denser and heavier in the periphery of these masses where coarse, dark blue granules of calcium are visible

in low power, and much lighter and finer granules are seen in the central parts of the masses, where fine dusting of the cytoplasm of the individual cells is visible under high power. The stroma consists of cellular fibrous tissue that contains a large number of blood capillaries and is in places infiltrated by small lymphocytes. Calcification of the stroma in the form of incrustation of its fibrous fibrils (which appear as dark blue wavy lines) is also present. Foreign body giant cells are found at the edges of the calcified masses and also free in the stroma. Characteristically shaped cholesterin crystals are found deposited both in the calcified epithelial masses and in the stroma. Typical bone tissue is found replacing the periphery of the calcified masses, varying from a very thin fringe to a thick deposit. Frequently such bone tissue includes calcified epithelial cells or contains marrow cavities filled with vascular fat tissue but without hematopoietic elements.

Discussion

From the description of our cases and of those reported in the literature it is clear that the calcified epitheliomas of the skin form a group of tumors that are clinically and pathologically characteristic. The benign behavior of the majority of them and their invariable tendency to undergo necrosis and calcification are the distinguishing features. They resemble the basal cell epitheliomas in many respects. Both consist of small, oval, deeply staining and closely packed cells, and both have the same type of distribution over the body, *i.e.* they are most frequently found over the head and neck. On the other hand, they are different in many important respects. The calcified epithelioma has a complete and well defined capsule, is freely movable and does not ulcerate or invade the overlying skin. The basal cell epithelioma has no capsule, is not freely movable but is locally invasive and produces a characteristic ragged ulcer (rodent ulcer). The cells of the calcified epithelioma rarely show mitosis and are grouped into round or elongated masses that are sharply demarcated from the stroma by a distinct basement membrane; the cells of the basal cell epithelioma, on the other hand, are apt to show frequent mitosis and grow in infiltrative, net-like (reticulated) strands. The cells of basal cell epithelioma only occasionally differentiate into squamous epithelium with cornification or pearl formation, whereas those of the calcified epithelioma almost

invariably differentiate in this manner. The calcified epithelioma always contains areas of necrosis and calcification, the basal cell epithelioma only rarely. The age incidence is different; the calcified epithelioma occurs in youth, the basal cell epithelioma occurs characteristically after 40. With basal cell epithelioma recurrence after extirpation is not uncommon; with calcified epithelioma it is extremely rare.

The rounded or ball-like contour and the subcutaneous position of the tumor with occasional traumatic ulceration of the overlying skin (following stretching by the expansile growth of the tumor) and discharge of necrotic epithelium have led to the clinical diagnosis of sebaceous (epidermoid) cyst, and the presence of bone tissue may cause the false impression of its being a dermoid or mixed tumor, but the anatomical structure is so different that it is superfluous to enter into a differential diagnosis here. The finding of typical squamous cells besides the small dark cells, and of the cornified pearls, should at once reveal to the observer the true nature of an epithelioma, although, when these characteristics are not evident, as in the totally necrotic or heavily calcified growths, confusion with other tumors, *e.g.*, endothelioma, sebaceous adenoma, and so on, may arise. The presence of fat in the epithelial cells and the deposit of cholesterin crystals in the necrotic epithelium have been taken, for instance by Eve, as an indication of the sebaceous origin of these tumors, but there is no ground for such an opinion since in any degenerating tissue the occurrence of fat and cholesterin is very common.

The different steps in the laying down of bone can be readily followed in our tumors. First the cell mass becomes necrotic, then calcified. The calcium is later replaced by new bone from the periphery inward. That the bone is formed largely as a result of ossification of the calcified masses is shown by the fact that the bony tissue has the same general shape or contour as the calcified masses, and that in the irregular beams, strands or trabeculae of bone tissue seen in the sections central inclusion of calcified epithelial cells is usually found, and also that all stages of transition from a calcified cell mass with just a narrow fringe of bone to one completely replaced by bone can be followed readily in the sections.

It is a very significant fact that 7 of our 9 cases (1 case with incomplete history not counted) are tumors which, although freely mov-

able over the deeper tissues, are adherent to the skin. Since these tumors are sharply encapsulated and do not invade the overlying skin, their adhesion to the latter can be explained only by the assumption that they are originally situated in the skin, or rather they are derived from the skin or its appendages.

There are certain features described in the literature that are not found in any of our cases and these are granulation tissue and myeloid tissue, obliterative changes in the arteries, which are looked upon as the cause of the necrosis, and myxomatous or sarcomatous degeneration of the stroma.

In 4 of our cases the peripheral portion of the tumor is definitely much more cellular and vascular than the central portion, and in 2 of these and 2 more from the rest of our cases the peripheral or subcapsular zone is moderately or densely infiltrated with lymphocytes and wandering cells. Similar findings have been made by Henzi.

Cystic spaces are found in the majority of our cases, and sometimes they are filled with fluid. These arise probably in the manner already mentioned above. Many of the spaces are irregular in shape and distribution and are empty and without fluid contents. Apparently they are artefacts.

As to whether trauma is the exciting cause of the tumor growth or not, the clinical histories of most of our cases do not give us satisfactory information. Only one case (Case 8) gives the previous history of a furuncle. It is possible in this case that during the healing of a furuncle the regenerating epithelium was displaced and later gave rise to a tumor. The common explanation of the traumatic origin of an epidermoid (sebaceous) cyst or calcified epithelioma, however, is that the mechanical injury dislocates or carries into the subepithelial tissue a bit of the epithelium that later grows into a tumor. It is hardly conceivable that trauma of this kind can be the chief cause, since these tumors are found not on the parts of the body that are more exposed to injury, but on the head and neck where trauma is relatively less frequent.

Although no good follow-up histories are available in our old operated cases, lack of recurrence after complete removal has been recognized in the literature as a feature characteristic of most of these tumors. Anatomically their non-malignant nature is evidenced by the constant possession of a well formed capsule, the

sharp demarcation of the epithelial masses and the scarcity of mitotic figures, together with the free-movability, the slow and expansile growth and the lack of ulceration. In view of the presence of cases of recurrence in the literature, however, the prognosis of these tumors should be guarded, even though the benign nature is apparent in most of them.

Summary

Ten cases of calcified epithelioma of the skin examined in the pathological laboratory of the Peiping Union Medical College are reported. These tumors form a distinct group of neoplasms that are anatomically and clinically well defined. They are circumscribed, well encapsulated growths beneath the skin, consisting of lobulated epithelial masses with a network of usually hyalinized fibrous stroma. The epithelial cells are small, oval, deeply staining and closely packed, and have a marked tendency to undergo necrosis, calcification and ossification. A study of our 10 cases and the 116 cases collected from the literature indicates that they are distributed most frequently on the head and neck and occur usually among the younger individuals. The large majority of these tumors are benign, but a few cases of recurrence following removal have been recorded.

The author wishes to express his appreciation to Dr. C. H. Hu for many valuable suggestions and aid in the preparation of the above article.

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DESCRIPTION OF PLATES

PLATE 72

FIG. 1. Cut surface of the tumor in Case 5. Natural size. Note its sandy or gritty appearance and the fact that the growth is enveloped by intact skin on one side and by a thin fibrous capsule on the other.

FIG. 2. Cut surface of the tumor in Case 7. Natural size. The tumor is subcutaneous, covered by skin on one side and by a thin fibrous tissue capsule on the other. Note the characteristic appearance of the cut surface.

FIG. 3. Tumor in Case 6. Natural size. Note relation of overlying skin to tumor.

FIG. 4. Tumor in Case 4. Natural size. Note encapsulation.

FIG. 5. Representative field of the tumor in Case 5, showing the general character of the growth. Note the alveolar arrangement of the epithelial masses. The living cell masses are stained deep blue by hematoxylin, appearing black in the picture. Practically all of them show either beginning or already extensive necrosis at the center. The necrotic epithelial masses are stained lightly with eosin and appear gray in the picture. × 13.

FIG. 6. A living epithelial cell mass from the tumor in Case 5. Note the small, oval, closely packed, deeply staining basal-like cells and their transition into squamous cells. × 155.

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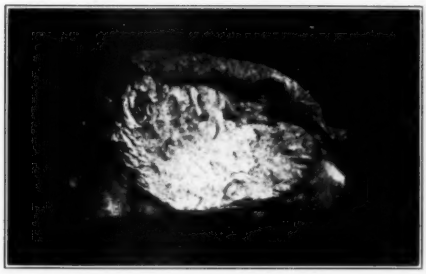

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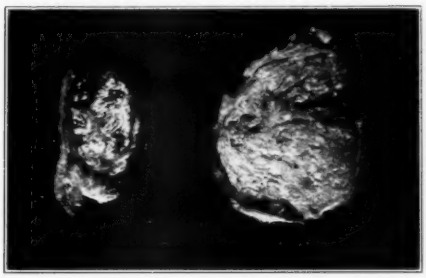

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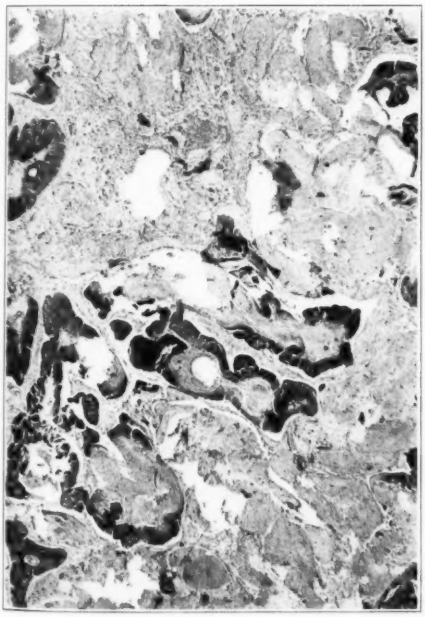

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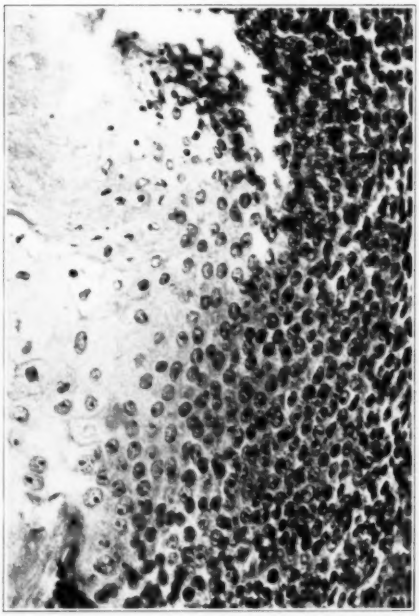

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Calcified Epithelioma of Skin

PLATE 73

FIG. 7. Epithelial pearl surrounded by a thick mantle of squamous cells. Basal-like cells at the periphery. × 155.

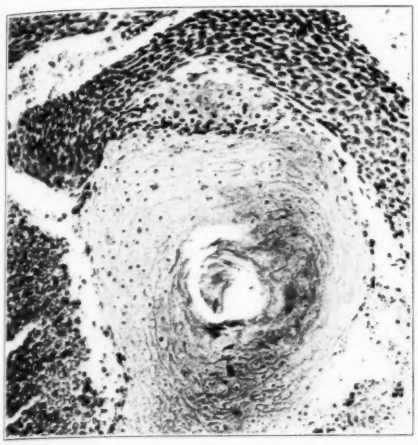
FIG. 8. Tumor in Case 5 showing giant cells at the periphery of a mass of degenerated squamous cells whose nuclei are no longer stained with hematoxylin but stained pink with eosin, yet whose structural details are well preserved. The stroma is slightly infiltrated with lymphocytes. × 155.

FIG. 9. Tumor in Case 5 showing the deeply staining areas of calcification in the degenerated epithelial masses and calcium incrustation of the fibers of the hyalinized stroma. × 70.

FIG. 10. Tumor in Case 2 showing spaces previously occupied by cholesterin crystals in the degenerated epithelial cell mass with large numbers of fine or coarse granules of calcium deposit giving a punctate appearance to the cells. × 155.

FIG. 11. Tumor in Case 4 showing calcium deposit in the totally degenerated epithelial masses and bone trabeculae, many of which include degenerated and calcified epithelium in their centers. No living epithelial masses are present in this tumor. Note the sharp demarcation of the tumor and its possession of a fibrous capsule at the right hand field. × 13.

FIG. 12. Tumor in Case 4, showing the ossification of a calcified epithelial mass from periphery inward. × 70.



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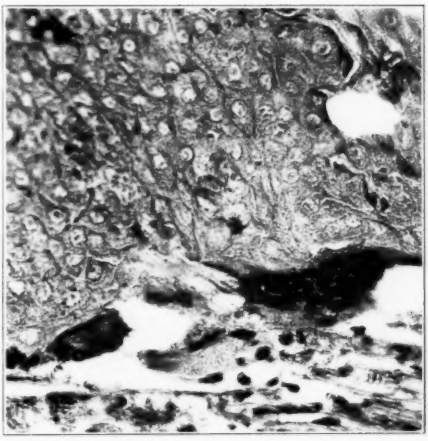

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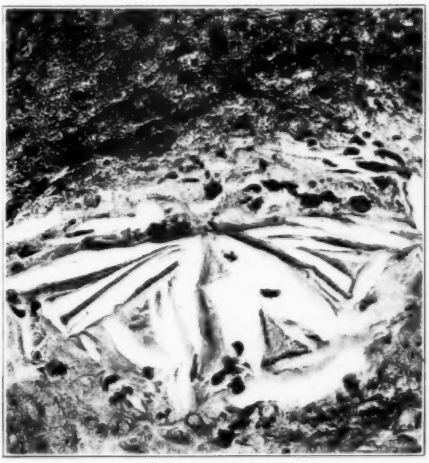

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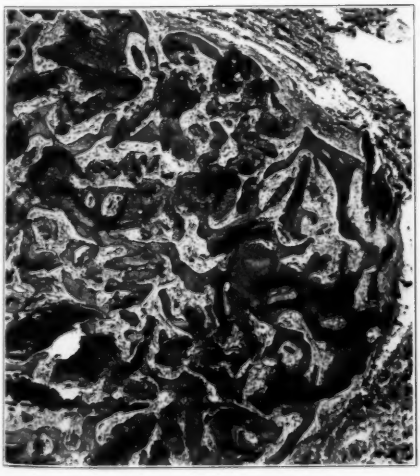

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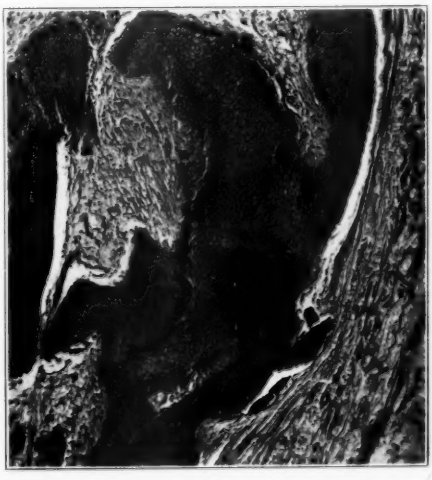

12

Calcified Epithelioma of Skin

SARCOMATOID METASTASES IN THE LYMPH NODES DRAINING A PRIMARY CARCINOMA WITH A SARCOMATOID STROMA *

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INTRODUCTION

The case I wish to report may be allotted to the category of the carcinosarcomas. I hesitate to do so for reasons that are made clearly evident in the text to follow. The remarkable point of interest in this case is, primarily, the interpretation of the sarcomatoid metastases in the lymph nodes draining a primary carcinoma with a sarcomatoid stroma.

The literature abounds with cases rightly and wrongly described as mixed tumors of the breast, thyroid and uterus. Occasionally, though rarely, two tumors are really present together, *i. e.* carcinoma and sarcoma. A true double tumor of such nature may be met with occasionally in the human subject. Great care must be exercised in interpreting appearances, as occasionally cancer cells may be diffusely arranged and even spindle-shaped. Cancer cells readily and frequently resemble mesoblastic elements, whether from pressure or from lack of differentiation, so that it is usually possible to raise a doubt as to whether the supposed sarcomatous elements are really such or merely deceptively altered epithelial cells.

I shall attempt to establish, therefore, that the sarcomatoid stroma in this case is not modified epithelium and that the spindle cells are of true connective tissue origin. This conclusion has been facilitated by the selective trichrome blue stain of Masson, and rechecked by Mallory's phosphotungstic acid hematoxylin stain in the histological study of the case.

Before describing this case I shall briefly review some of the controversies held on the subject of mixed carcinosarcomas. The state of utter confusion and the various opinions existing permit this further study.

* Received for publication March 4, 1933.

We are not concerned in this discussion with the primary mixed tumors. That certain tumors originate through neoplastic growth of both stroma and epithelial cells in a tissue complex is conceded. It is difficult to determine which of the carcinosarcomas originate in this manner.

We are concerned here with secondary mixed tumors, that is, sarcoma arising from the stroma of carcinoma. This, of course, is not universally tenable. Several theories have been advanced, but the main schools of thought are:

(*a*) Sarcomatous transformation of the connective tissue stroma of carcinoma (Haaland,[1] Ehrlich[2]).

(*b*) Spindle cell metaplasia of the epithelial cells. The altered epithelium may simulate a sarcoma in appearance (Ewing[3]).

(*c*) These sarcomas are nothing but an exuberant proliferative growth of ordinary connective granulation tissue (Orth[4]).

It was a most surprisingly new fact, discovered by Ehrlich and Apolant,[5] that the stroma in certain transplantable mouse carcinomas gave rise to a new and distinct kind of tumor tissue, *i. e.* a transplantable sarcoma. In succeeding generations the sarcoma ousts the carcinomatous elements and entirely supplants them. Ultimately a pure sarcoma is obtained. This phenomenon has been observed by many others.

Haaland agrees with the opinions of Ehrlich and Apolant that the development of sarcoma is explicable as a new or altered form of specific reaction to influences proceeding from the carcinoma cells. This enhanced reaction on the part of the connective tissue, progressing to sarcoma, depends not only on the stimulating influence proceeding from the carcinoma but also on the individual response of the host.

The question arises whether these sarcoma-like tumors are true malignant newgrowths and not mere infectious granulomas or granulation tissue around the carcinoma alveoli. Orth, commenting on Lewin's case,[6] questioned the existence of sarcoma. He thought sarcoma was not really present but rather a growth of connective granulation tissue. The presence here and there of spindle cells is also a feature of ordinary proliferations of connective tissue.

Ehrlich and Apolant rejected the possibility of the new tumors being granulomas since metastases in the pulmonary artery by

emboli of tumor cells continued growing. The presence of collagen fibrils between the cells in intravascular secondary nodules proved the connective tissue origin of the cells.

Murray [7] has strongly substantiated the belief that sarcoma arises from the stroma and not from altered cancer cells. Haaland, under high magnification, observed in the cytoplasm of these rapidly growing, spindle-shaped cells characteristic fine fibrils (Mallory's fibroglia fibrils) like those found in young connective tissue (Maximow [8]). Other fibrils were found outside the cells, giving all the reactions of collagen. The production of collagenous fibrils and the presence of fibroglia fibrils in the cells proved their origin from fibroblastic elements of the connective tissue. In my opinion this justifies calling these tumors true sarcomas. Murray dismisses the idea of the possibility of the sarcomatous cells being derived from carcinoma parenchyma since these cells present all the characteristics of true connective tissue cells as described above. There exists no analogy for a process of metaplasia that can transform typical epithelial cells into equally typical connective tissue cells. It is, however, generally admitted that the epithelial cells of the neuroglia have the faculty of producing a network of fibrils.

Dorsch [9] in 1896 reported a case of carcinosarcoma. He concluded that sarcoma arose through stimulation of connective tissue by the epithelial cells. This curious transformation of the stroma in cancers has been observed by many others. Oertel [10] classifies carcinosarcomas into several categories, but says that the "genuine carcinosarcoma is the sarcoma that develops out of the stroma of an already existing cancer." The stroma acquires new characteristics and begins to proliferate. Occasionally it assumes neoplastic characteristics. In this manner carcinosarcoma results (Masson [11]). Mallory [12] favors the view that in epithelial tumors (especially skin and mammary gland) the cells can chemically or otherwise directly influence and determine the amount of the stroma. The growth is at times so active that it is not unusual to find fibroblasts in mitosis proliferating actively and producing many collagen fibers.

On the other hand Ewing (in answer to Ehrlich and Apolant, Haaland, and others) finds it difficult to conceive how an original carcinoma can be made to yield all its growth energy to normal stroma cells and completely retire from the field, yet this anomalous result has been reached in the pure sarcoma developing from mouse

carcinoma. In lower animals the ready response of fibroblasts to various stimuli renders more acceptable the belief that transplanted stroma occasionally assumes neoplastic properties during a series of transfers, but does not prove the existence of such a process in the course of tumor growth in man. Ewing believes that the influence of transplantation renders these observations inapplicable to human tumors and that several uncertainties still surround the interpretation of this remarkable change in structure.

Ewing feels the correct interpretation of the carcinosarcoma structures in man and lower animals is a matter of difficulty. The chief source of these structures is the transformation of epithelial cells into spindle cells. This change is of widespread occurrence in epithelial tumors, and is facilitated by rapid growth, inflammatory exudate and relief from pressure. He has seen pure spindle cell metastases in epithelioma of the lip, adamantinoma, and so on. Krompecher [13] believes that the carcinosarcoma is merely a carcinoma, its spindle-shaped cells being epithelial in spite of its morphological resemblance to fibroblasts. His studies on the metaplasia of squamous epithelium have an important bearing on this subject.

On examining several of Woglom's cases purporting to show the sarcomatous transformation of stroma cells Ewing concluded that the spindle cells were derivatives of epithelium. This view has been enthusiastically accepted by many. Gessner [14] refers to all so-called carcinosarcomas of the uterus as epithelial in origin. Meyer [15] believes that sarcomatous transformation of carcinomas has not been demonstrated.

Helwig [16] reviews twenty-four cases of reported mixed tumors of the breast and concludes that it is impossible to draw any definite conclusions as to whether the tumors are true carcinosarcomas, or if metaplasia has occurred with a gradual transition from a carcinoma into a sarcoma-like structure. In his own case, which simulated histologically a picture of a carcinoma and giant cell sarcoma, he is convinced that it is a carcinoma with a sarcomatous metaplasia of epithelial cells.

Out of a considerable number of so-called mixed tumors of the uterus Herxheimer [17] found only five that could withstand critical analysis. Schmorl's case of thyroid adenoma, recurring as carcinoma with sarcomatous stroma and with sarcomatous metastases,[18] is

doubted by Ewing. In thyroid tumors the form of epithelial cells is subject to wide variations, a fact that renders Schmorl's interpretation of his carcinosarcoma very questionable. Definite association of sarcoma with carcinoma of the breast has been reported by a few observers, but in several such cases Ewing has not been satisfied that the spindle cell areas were not modified epithelium. Nevertheless, Mason and Wells,[19] in reporting their case of carcinosarcoma, assert that it is not a carcinoma with altered epithelial cells since the carcinoma portion metastasized to the cervical lymph nodes and the sarcoma portion to the kidney.

Indeed, many observers agree that in rare cases we do not know whether a given cell group originates in epithelium or not, since the facts do not justify such definite distinctions between epithelial and certain other tumors. Cohn,[20] in reviewing this subject, maintains that our available means for differentiating epithelial and sarcomatous elements are still so defective that a positive proof cannot usually be offered to support a diagnosis of mixed sarcoma and carcinoma, especially in cases of advanced spontaneous tumors.

Finally, it is to be remembered that in some forms of basal cell carcinoma the epithelial cells may have an indifferent spindle-shaped contour, often elongated, even fibrillar (Kaufmann[21]). Cancers arising from hair matrices often show large masses of spindle cells running in bundles, frequently produce numerous long and coarse fibrils, and may suggest spindle cell sarcoma (Mallory[12]). Kaufmann,[21] in quoting Krompecher, emphasizes the possible variability of ectodermal cell proliferation, especially the transition of ectodermal into apparently mesenchymal structures. Masson[22] in his Atlas of Cancer shows beautifully colored illustrations of mixed tumors of the parotid in which the ectodermal tumor cells demonstrate their capacity to assume the appearance of mesenchymal tissue.

In view of the interest in these tumors the following case is reported.

Report of Case

Cliincal History: Mrs. A. L., aged 74 years, was admitted to the Notre Dame Hospital Dec. 15, 1931 with a superficial, ulcerated tumor 3 cm. by 2.5 cm., situated in the skin of the left mammary fold, medial to the nipple line and definitely independent of the mammary gland itself. It was quite hard and had

an inflammatory zone about it. The tumor was removed and sections made for histological study. A diagnosis of carcinosarcoma was made.

At that time lymph nodes in the axilla were not palpable. Radical excision of the breast was advised but the patient refused. The biopsy incision healed satisfactorily and the patient left the hospital in good condition. Physical examination revealed nothing of importance.

Two months later the patient noticed a swelling in the left axilla. This gradually enlarged and she finally returned to the hospital Sept. 9, 1932 (almost eight months later). At operation one lymph node 2.5 by 1 cm. in size and another 1.5 by 1.5 cm. were removed and sections made for histological study. The lymph nodes were diagnosed as fibrosarcoma.

The patient has been under observation and has been well and free from further recurrences.

Microscopic Description of the Primary Tumor

The tumor is spherical in form and for descriptive purposes it may be divided into three zones of a circle.

Outer Zone: Epithelioid processes growing centrifugally from the center of the tumor invade the epidermis and the underlying dermis. The epidermis is locally ulcerated. The cancer growth is of the higher type, preserving some of the attributes of its epithelial origin. A few prickle cells can be seen in some of the strands invading the stroma. Here and there an abortive attempt at cornification by individual cells is observed. The cancer cells are polygonal in shape, well formed, evenly stained, showing relatively few mitoses. Most of the lymphatics are plugged with large epithelioid processes growing transversely, although some of the lymphatics are empty and free. The connective tissue stroma varies considerably in different areas. For the greater part the stroma is quite delicate and not very cellular. Toward the periphery at each end the connective tissue is quite mature, displaying very few cells and a well stained collagenous material. At other points the stroma forms a loose connective tissue of large cells regularly ensheathing the epithelial strands and encasing empty lymphatics. There is a marked inflammatory reaction, particularly near the ulcerated zone, in the form of many plasma cells, some lymphocytes and a few eosinophils. The blood vessels are free from invasion, although a few show endothelial proliferation within the lumen.

Mid Zone: The epithelial strands are not as numerous as in the outer zone, and show some parenchymal degeneration. Accompanying the degeneration within these strands is a markedly abundant

stroma that has become frankly sarcomatoid in character. It now shows increased cellularity and numerous cell divisions. The cells are larger and usually elongated, though varying in shape. This rapidly proliferating connective tissue forms broad bands of spindle-shaped cells surrounding the cancer alveoli (almost in halo fashion) and separates them from one another. In other areas of this zone a further and greater activity of the stroma is observed. Here, the cellularity and proliferating quality of the stroma is sarcomatous in nature. The cells are large, rapidly dividing and show abundant fibroglia fibrils and collagenous fibrils between the cells.

The fibroblastic proliferation is here characterized by an immature manner of growth, proliferating without restraint, showing a number of monstrous cells with bizarre mitotic and multipolar figures and nuclei of extraordinary size. Hyperchromatic and hypochromatic nuclei are frequent. The amount of cytoplasm varies greatly and a marked polymorphism prevails. The cells are much larger than usually seen in granulation tissue. Here the stroma reaction is so intense that it overrides and eats into the cords of cancer cells so that within this stroma only scattered epithelial cells are observed. These epithelial cells are unlike the neighboring stroma cells which are pale, irregular in shape and contour, and with definite fibroglia fibrils (as seen by the trichrome stain).

Central Zone: The epithelial strands are very much necrosed. The collagen fibrils laid down by the fibroblasts are thick, well stained and sclerotic. Atrophic and necrotic changes are observed here in the epithelial alveoli surrounded by this sclerotic tissue. These changes proceed from the center of each alveolus toward its periphery, often leaving only a single layer of cancer cells close to the stroma tissue, while the rest of the alveolus is necrotic. In the center the epithelial strands are almost wholly sclerosed with calcareous deposits in evidence. The stroma has become hyalinized and acellular.

Microscopic Description of the Lymph Nodes

In the larger lymph node at one end there still remains the normal structure of lymphoid tissue. Everywhere else a fibrosarcomatous tissue replaces the lymph node tissue and its capsule, and also invades the pericapsular fat. The hilum seems to have been first at-

tacked. The areas neighboring the hilum are sclerotic. The direction of the growth of the tumor is from within outward, following the sinuses. There seems to be a transformation of endothelial cells into fibroblasts *in situ*. Epithelial strands, or cells resembling those of the primary tumor, cannot be found but mixed among this sarcomatoid structure are many polymorphous cells. The origin of these large polymorphous cells is probably from the mesenchymal tissue of the lymph node.

At one point in the periphery of the lymph node, an afferent lymphatic vessel is seen, within which free epithelial cells are floating in the lymph. This is the only definite trace of epithelial cells, but this is evidently a later stage of the picture. The cells lie free and unattached, unable to enter the lymph node, the lymphatics of which have been blocked by the fibrosarcomatous replacement of the tissue. Most of the other lymphatics are empty, some are filled with lymph, while others show a marked endothelial proliferation. The blood vessels do not show any involvement other than endothelial proliferation. For the greater part this fibrosarcomatous tumor consists of interlacing bundles of spindle cells. Between the individual cells fine collagen fibrils are to be seen. With higher magnification fine fibrils are found in the cytoplasm of the cells (fibroglia fibrils, Mallory).

The smaller nodule, round in shape, shows traces of its lymphoid structure. The lymph node is practically completely replaced by the fibrosarcomatous tissue. The capsule is entirely destroyed and the surrounding fat is invaded to a greater degree than in the one previously described.

Discussion

1. These spindle-shaped cells are not the result of epithelial metaplasia. Transition figures between these cells and cancerous epithelial cells are not to be found. Frequently many degenerating cancer cells isolated in the sarcomatoid stroma are seen. Sarcomatous transformation of the stroma cells results (*a*) when in contact with epithelial cells which, moreover, are degenerating or dead, and (*b*) when surrounding lymphatics, whether they contain epithelial cells or not.

It appears that the transformation and the multiplication of the stroma cells are determined by some substance contained by the

cancer cells. When their degeneration occurs this substance is liberated and acts directly on the fibroblasts whether it be *in situ* or after being absorbed by the lymphatics. As we shall see later, this is the only explanation that allows us to understand the sarcomatoid metastases in the lymph nodes, their structure and manner of growth. If these spindle-shaped cells are but altered epithelial cells their appearance in the lymph nodes is not altogether surprising. It would be difficult to explain the fact that the lymphatics in the primary tumor were plugged with well stained, polygonal-shaped epithelial cells, and that nowhere could a lymphatic vessel be found containing spindle-shaped cells.

Histologically these spindle-shaped cells behave like fibroblasts and possess the same selective staining properties. Even if the general appearance and behavior of the rapidly growing spindle-shaped cells would not allow any conclusions as to their origin, other points might permit a decision as to which elements they have arisen from. Using Masson's trichrome blue technique, fibroglia fibrils stain red and collagen fibers stain blue. Under high magnification characteristic, fine, red fibrils are seen in the cytoplasm, like those described by Mallory and similar to the fibroglia found in young connective tissue (rechecked by Mallory's phosphotungstic acid hematoxylin). These red fibrils are found intracellularly following the prolongations of the cell; other such fibrils are found extracellularly. Between the cells collagen fibrils staining blue are numerous. The cells behave like fibroblastic tissue, forming a scaffold of interlacing bundles of spindle cells. As the collagenous material laid down by these cells becomes excessive the cells themselves become scarce.

The presence of fibroglia fibrils in the cells, the production of collagenous fibrils outside the cells and their behavior, aside from their general appearance, prove their origin from fibroblastic elements of the connective tissue. It is, therefore, justifiable to dismiss the possibility of epithelial metaplasia and to suggest an explanation as to the histogenesis of the spindle cells.

2. It is necessary, in dealing with mixed tumors, to establish whether they are primarily or secondarily mixed, as previously pointed out. Here the primary skin tumor is not primarily a mixed tumor. The cancer and sarcomatous stroma are not proliferating together. The cancer only invades and provokes the production of a stroma at the expense of the local connective tissue. The tumor is

secondarily mixed. It is only at some distance from the zone of invasion that the stroma assumes sarcomatous characteristics.

3. How can the appearance of fibrosarcomatous tissue in the lymph nodes be accounted for in this case? Generally, sarcomas do not metastasize along lymphatics. Lymphosarcomas are the only sarcomas that frequently travel by the lymphatics and the origin of these tumors within the lymph nodes fully accounts for this tendency. Melanosarcomas are epithelial in origin (Masson), which accounts for their specificity in affecting lymph nodes. Most statistical reports of lymph node invasion in sarcoma are of doubtful value owing to the uncertainty connected with the diagnosis of sarcoma. Most of the tumors reported as sarcomas of the testis and thyroid are probably epithelial tumors and commonly invade the lymph nodes (Ewing).

On the other hand it has been observed that sarcomas, although having metastasized by the blood stream, nevertheless set up irritative endothelial proliferation in the neighboring lymph nodes. Usually the soluble products absorbed by the lymphatics from neoplastic nests (cancerous or sarcomatous) initiate only an endothelial hyperplasia, followed by fibroblastic transformation and sclerosis. In this case the stroma cells of the lymph node bathed in the lymph that had absorbed the products liberated by the epithelial cells in the primary tumor. A sarcomatoid transformation of these stroma cells resulted (just as there was a sarcomatoid transformation of the stroma around the lymphatics at the edge of the primary tumor).

4. And lastly it is necessary to prove that in the lymph nodes small remains of carcinoma still were not present, inciting the connective tissue to an actively proliferative, granulomatous growth, thereby explaining the appearance of a presumably sarcomatoid tissue in the lymph node.

Metastases of cancer growth can lead to an abundant stroma corresponding more or less closely in amount to the original tumor, even in a tissue containing as little connective tissue as the bone marrow. The behavior of the stroma may become so active that it is often difficult to distinguish sharply between those tumors whose stroma is clearly not neoplastic and those in which it constitutes a part of the tumor and which must be regarded as mixed tumors. In this case there may have been an epithelial metastasis in the lymph node with a resulting sarcomatoid transformation of the stroma. De-

generation of the cancer cells followed, allowing the sarcomatoid tissue alone to persist and to proliferate actively. This is possible, but not probable. That the products liberated by the degenerated epithelial cells in the primary tumor were absorbed by the lymphatics and directly influenced the stroma reaction on reaching the nodes is the preferable view-point. This point is important. In the irritative endothelial hyperplasia of lymph nodes the toxic products entering the afferent lymph vessels of the nodule drain along the sinuses and reach the hilum. These products first initiate this endothelial hyperplasia at the hilum; this reaction then radiates outwardly toward the cortex, if the irritative stimulus is great or prolonged. In this case a similar process has occurred (the hilum having been first attacked, as previously pointed out in the microscopic description of the lymph nodes) with not only a fibroblastic transformation of the endothelial cells lining the sinuses, but also a sarcomatoid transformation. This is what has occurred here, rather than a neoplastic invasion, for if this had been so, evidence would exist in the cortex of the lymph node near the entrance of the afferent lymph vessels. The changes in the lymph node in this case are comparable to the usual irritative endothelial hyperplasia. This permits the belief that neither an epithelial nor a sarcomatous metastasis has occurred, but rather a sarcomatoid transformation *in situ* similar to that occurring around the cancer alveoli of the primary tumor.

It will be interesting to know if this sarcomatoid tissue of the primary tumor and the lymph gland is truly sarcomatous, that is to say, now possesses the autonomy of newgrowth with its characteristic independent proliferative qualities, or if it is only sarcomatous in appearance, due to especially irritative products originating from the cancer cells. In this regard the further course of the disease alone can inform us. If the tumor is truly sarcomatous, then metastasizing by way of the blood stream and not lymphatics is probable (*cf.* case of Vadon *). If the tumor is pseudosarcomatous, the appearance of new fibroblastic tumors will be dependent on carcinomatous metastases or a local recurrence.

* Professor Masson relates the following incident concerning Vadon, an interne in one of the hospitals of Paris. While aspirating the fluid from a cystic mass of the breast he accidentally stuck his hand with the aspirating needle. The mass proved to be carcinoma at later examination. In the meantime a growth in his hand developed at the point of injury. This growth was purely fibrosarcomatous. This generalized by way of the blood stream and he died. The metastases proved to be pure sarcoma.

Summary and Conclusions

1. A case of sarcomatoid metastases in lymph nodes draining a primary carcinoma with a sarcomatoid stroma is reported.

2. The usefulness of Masson's trichrome blue stain, particularly in the histological study of the fibroglia of fibroblasts, is here emphasized.

3. The possibility of epithelial metaplasia is discussed.

4. An analogy between this case and the experimental tumors of Ehrlich, Haaland, and others is assumed.

5. The dividing line between mixed tumors and tumors whose stroma is not clearly neoplastic is difficult to determine. In this case further metastases by way of the blood stream alone will decide.

6. The occurrence of sarcoma in the lymph nodes in this case may be explained on the basis of absorption of products from degenerated epithelium by the lymphatics. This initiates a stroma reaction in the lymph nodes which may become neoplastic in character, depending on the tissue response of the host and the irritative quality of the absorbed products liberated by the degenerating cancer cells.

I am indebted to Professor Masson for his kindly interest, guidance and encouragement in this work, and I am also indebted to Dr. L. C. Simard for the preparation of the photomicrographs.

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DESCRIPTION OF PLATE

PLATE 74

FIG. 1. Primary tumor. Carcinomatous plug of epithelioid cells. The surrounding connective tissue stroma is quite normal. Note the marked inflammatory stroma reaction. × 600.

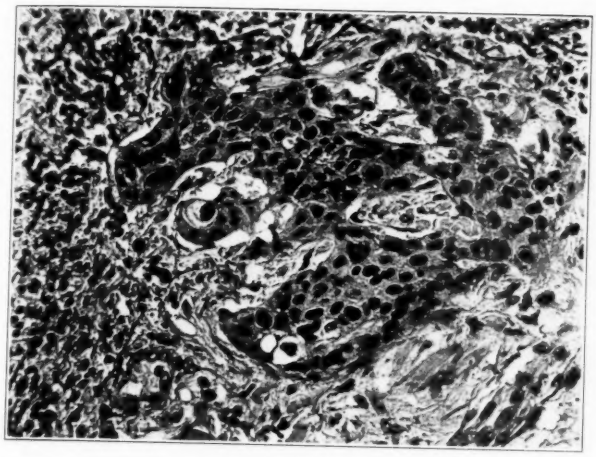
FIG. 2. Primary tumor. One of the cancer plugs is completely necrosed. The connective tissue stroma is active, proliferating and sarcomatoid in nature. In the center a few mitotic figures are seen; one of these shows abnormal mitosis. × 600.

FIG. 3. Lymph node. The lymphoid tissue is replaced by a tissue resembling the sarcomatoid stroma surrounding the necrotic cancer plug in Fig. 2.

At left, spindle-shaped cells cut longitudinally.

On the right, most of the cells cut transversely. Lower right, a few mitotic figures. Upper right, a large hyperchromatic nucleus is seen. The fine intercellular stroma and collagenous fibrils are visible. × 600.

Masson's hemalum-erythrosine-saffron stain is the routine stain used in our laboratory.



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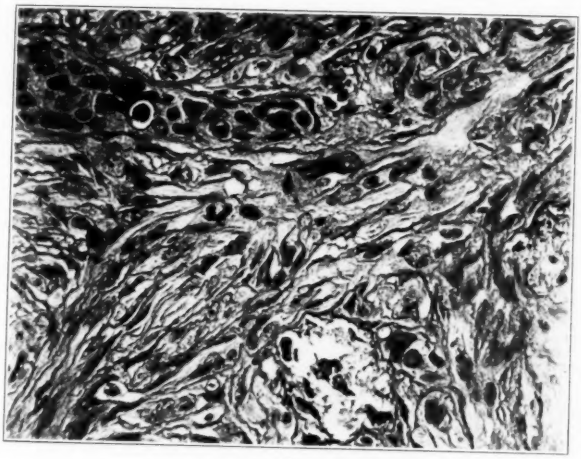

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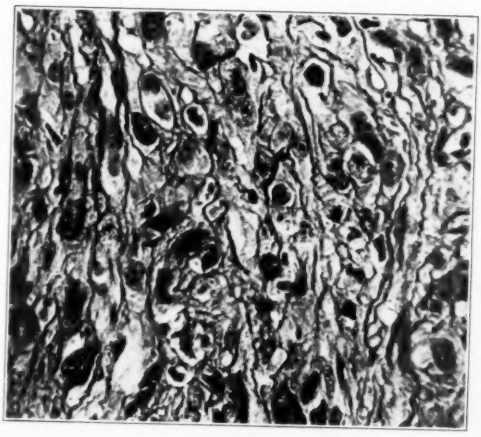

3

Greenblatt

Sarcomatoid Metastases in Lymph Nodes